DRAFT

Water Quality Criteria Report for Trifluralin

Phase III: Application of the pesticide water quality criteria methodology



Prepared for the Central Valley Regional Water Quality Control Board

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June 2016

Disclaimer

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List of acronyms and abbreviations

AF Assessment factor

APHA American Public Health Association

ASTM American Society for Testing and Materials

BAF Bioaccumulation Factor
BCF Bioconcentration Factor
BMF Biomagnification Factor
CAS Chemical Abstract Service

CDFG California Department of Fish and Game

CSIRO Commonwealth Scientific and Industrial Research Organization, Australia

CVRWQCB Central Valley Regional Water Quality Control Board

DPR Department of Pesticide Regulation

EC_x Concentration that affects x% of exposed organisms

FDA Food and Drug Administration

FT Flow-through test

 IC_x Inhibition concentration; concentration causing x% inhibition

ICE Interspecies Correlation Estimation

IUPAC International Union of Pure and Applied Chemistry

K Interaction Coefficient K_H Henry's law constant

 K_{ow} Octanol-Water partition coefficient K_p or K_d Solid-Water partition coefficient

 LC_x Concentration lethal to x% of exposed organisms

 LD_x Dose lethal to x% of exposed organisms

LL Less relevant, Less reliable study

LOEC Lowest-Observed Effect Concentration

LR Less relevant, Reliable study

MATC Maximum Acceptable Toxicant Concentration

Not relevant or Not reliable study

n/a Not applicable

NOEC No-Observed Effect Concentration

NR Not reported

OECD Organization for Economic Co-operation and Development

pK_a Acid dissociation constant RL Relevant, Less reliable study RR Relevant and Reliable study

S Static test

SMAV Species Mean Acute Value

SR Static renewal test

SSD Species Sensitivity Distribution
TES Threatened and Endangered Species

US United States

USEPA United States Environmental Protection Agency

1 Introduction

A methodology for deriving freshwater water quality criteria for the protection of aquatic life was developed by the University of California - Davis (TenBrook et al. 2009a). The need for a methodology was identified by the California Central Valley Regional Water Quality Control Board (CVRWQCB 2006) and findings from a review of existing methodologies (TenBrook & Tjeerdema 2006, TenBrook et al. 2009b). The UC-Davis methodology is currently being used to derive aquatic life criteria for several pesticides of particular concern in the Sacramento River and San Joaquin River watersheds. The methodology report (TenBrook et al. 2009a) contains an introduction (Chapter 1); the rationale of the selection of specific methods (Chapter 2); detailed procedure for criteria derivation (Chapter 3); and a criteria report for a specific pesticide (Chapter 4). This criteria report for trifluralin describes, section by section, the procedures used to derive criteria according to the UC-Davis methodology. Also included are references to specific sections of the methodology procedure detailed in Chapter 3 of the report so that the reader can refer to the report for further details (TenBrook et al. 2009a).

2 Basic information

Chemical: Trifluralin (Fig. 1)

CAS: Benzenamine, 2,6-dinitro-N,N-dipropyl-4-(trifluoromethyl)-

CAS Number: 1582-09-8 USEPA PC Code: 036101 CA DPR Chem Code: 597

IUPAC: α,α,α-trifluoro-2,6-dinitro-N,N-dipropyl-p-toluidine

Chemical Formula: C₁₃H₁₆F₃N₃O₄

$$H_3C$$
 O_2N
 NO_2
 CF_3

Figure 1 Structure of trifluralin

(source: USEPA 1996)

Trade names: Treflan, L-36352, Crisalin, Su Seguro Carpidor, Trefanocide, Treficon, TR-10, Triflurex, Trim, Ipersan, Sinflouran, Ipifluor, Flurene SE, Tri-4, Trust, M.T.F., Trifluralina 600, Elancolan, Su Seguro Carpidor, Trefanocide, Treficon, and Ipersan

3 Physical-chemical data

Molecular Weight

335.279

(http://webbook.nist.gov/cgi/inchi/InChI%3D1S/C13H16F3N3O4/c1-3-5-17(6-4-2)12-10(18(20)21)7-9(13(14%2C15)16)8-14(12)16(20)227.7 ph/9/262 (M20)261 (M2)

11(12)19(22)23/h7-8H%2C3-6H2%2C1-2H3)

Density

1.3 g/mL (PPDB 2016)

Water Solubility

0.3 mg/L at unknown temperature (Hornsby et al. 1996.)

0.3 mg/L at unknown temperature (WSSA 1989)

0.221 mg/L at unknown temperature (Tomlin 1997)

0.209 mg/L at unknown temperature (USEPA 2015)

0.184 mg/L at unknown temperature (USEPA 2015)

Geometric mean: 0.238 mg/L

Melting Point

49°C (USEPA 2015) 42-49°C (USEPA 1996) 46-47°C (EXTOXNET 2016)

Geometric mean: 45.9°C

Vapor Pressure

0.0018 Pa at 25°C (USEPA 2015) 0.0095 Pa at 25°C (PPDB 2016) **Geometric mean:** 0.014 Pa 25 °C

Henry's constant (K_H)

2.12 x 10 ⁻⁴ Pa m³ mol⁻¹ (USEPA 2015) 1.03 x 10 ⁻⁴ Pa m³ mol⁻¹ (USEPA 2015) 1.5 x 10 ⁻⁴ Pa m³ mol⁻¹ (Day 1987) **Geometric mean:** 1.3 ⁻⁴ Pa m³ mol⁻¹

Organic Carbon Sorption Partition Coefficients (log K_{oc})

All values from USEPA 2015

4.215 4.252

Geometric mean: 4.233

$Log K_{ow}$

*Values referenced from the BioByte Bio-Loom program (2015)

- 5.31 (USEPA 2015)
- 5.34 (USEPA 2015)
- 3.06 (Metcalf, *no date**)
- 5.28 (Brown and Flagg 1981*)
- 4.88 (Saito et al. 1993*)
- 3.97 (Kanazawa 1981*)
- 4.82 (Finizio et al. 1997 *)
- 4.98 (Donovan and Pescatore 2002*)

Geometric mean: 4.64

Bioconcentration Factor

Table 1 Bioconcentration factors (BCF) for trifluralin

FT: flow-through, SR: static renewal, S: static, NR: not reported; values are on a wet weight basis and are not lipid-normalized.

Species	BCF	Exposure	Reference
NR	202.4	NR	USEPA 2015
NR	321	NR	PPDB 2016
NR	207.6	NR	USEPA 2015
Oncorhynchus	13,000	S	Schultz 1999
mykiss			
Lepomis	5,304	S	Schultz 1999
macrochirus			
(16°C)			
Lepomis	15,506	S	Schultz 1999
macrochirus			
(23°C)			
Ictalurus	5,870	S	Schultz 1999
punctatus (23°C)			
Ictalurus	2,258	S	Schultz 1999
punctatus (16°C)			
Micropterus	1,681	S	Schultz 1999
salmoides			
Dorosoma	8,912	S	Schultz 1999
cepedianum			
Acipenser	419	S	Schultz 1999
fulvescens			
	GEOMEAN		·
	1905.11		

Environmental Fate

Table 2 Trifluralin hydrolysis and photolysis and other degradation.

(NR: not reported).

(NR: not reported	u).				
	Half- life (h or d)	Water	Temp (°C)	pН	Reference
Hydrolysis	1199 d	Aqueous buffer	22	4.1	Ramesh 1999
	1029 h	Aqueous buffer	22	7.1	Ramesh 1999
	774 h	Aqueous buffer	22	9.1	Ramesh 1999
Aqueous Photolysis	8.93 h	Aqueous buffer	25	7.0	Carpenter 1988
	0.2 h	Distilled	25	5.35	Dimou 2004
	0.4 h	Seawater, 33.4 % salinity	25	7.62	Dimou 2004
	0.49 h	River water	25	7.81	Dimou 2004
	0.84 h	Lake water	25	7.87	Dimou 2004
Biodegradation (aerobic)	128 d	Silty clay	25	NR	Tiryaki 2004
	98 d	Clay	25	NR	Tiryaki 2004
	126 d	Clay	25	NR	Tiryaki 2004

4 Human and wildlife dietary values

There are no FDA action levels for trifluralin in food (USFDA 2000) and there are no EPA pesticide tolerances set for any aquatic species (USEPA 2012).

Wildlife LC₅₀ values (dietary) for animals with significant food sources in water

The US EPA Environmental Risk Assessment for the Reregistration of Trifluralin (USEPA 1996) states that trifluralin is practically nontoxic to birds for acute and subacute exposures. The reported acute oral LC_{50} for mallard exceeds 2,000 mg/kg (Hudson et al. 1984) and the subacute dietary LC_{50} exceeds 5,000 mg/kg. The latter study was not available for evaluation from the US EPA.

No LC₅₀ data was available for wildlife species with significant food sources in water during the present report preparation. If highly rated measured data for mallard duck become available in the future, they should be examined to determine the potential risk to wildlife.

Wildlife dietary NOEC values for animals with significant food sources in water

The Reregistration report (USEPA 1996) reports a NOEC value of 910.5 mg/kg with the caveat that the value is based on unreviewed data. This study was received from the US EPA under MRID 40334704 and rated highly (Beavers et al. 1987). No other NOEC data was available for wildlife species with significant food sources in water during the present report preparation. If highly rated measured data for mallard duck become available in the future, they should be examined to determine the potential risk to wildlife.

5 Ecotoxicity data

Approximately 54 original studies on the effects of trifluralin on aquatic life were identified and reviewed. In the review process, many parameters were rated for documentation and acceptability for each study, including, but not limited to: organism source and care, control description and response, chemical purity, concentrations tested, water quality conditions, and statistical methods (see Tables 3.6, 3.7, 3.8 in TenBrook et al. 2009a). Single-species effects studies that were rated as relevant (R) or less relevant (L) according to the method (Table 3.6) were summarized in data summary sheets. Information in these summaries was used to evaluate each study for reliability, using the rating systems described in the methodology (Tables 3.7 and 3.8, section 3-2.2, TenBrook et al. 2009a), to give a reliability rating of reliable (R), less reliable (L), or not reliable (N).

Studies of the effects of trifluralin on mallard ducks were rated for reliability using the terrestrial wildlife evaluation. Mallard studies rated as reliable (R) or less reliable (L) were used to consider bioaccumulation. One study for mallard duck rating R was located in the literature and it is summarized in Section 4.

Copies of completed summaries for all aquatic studies are included in the Appendix of this report. All data rated as acceptable (RR) or supplemental (RL, LR, LL) for criteria derivation are summarized in Tables 3 - 10, found at the end of this report. Acceptable studies rated as RR are used for numeric criteria derivation, while supplemental studies rated as RL, LR or LL are used for evaluation of the criteria to check that they are protective of particularly sensitive species and threatened and endangered species. These considerations are reviewed in section 10.1 and 10.3 of this report, respectively. Studies that were rated not relevant (N) or not reliable (RN or LN) were not used for criteria derivation.

No acceptable microcosm studies were identified in the literature.

Evaluation of aquatic animal data

Using the data evaluation criteria (section 3-2.2, TenBrook et al. 2009a), three acute studies yielding four toxicity values from three taxa were judged reliable and

relevant for acute criterion derivation (Tables 3-4). Seven acute toxicity animal values for seven taxa from three studies were rated RL, LL, or LR and were used as supplemental information for evaluation of the derived acute criteria in the Sensitive Species section 10.1 (Table 5). Four chronic animal toxicity values from four studies were rated RR (Tables 7-8). One chronic toxicity animal value from one study was rated RL, LL, or LR (Table 10).

Evaluation of aquatic plant data

Plant data were used to derive the chronic criterion instead of chronic animal data because trifluralin is an herbicide and plants are the most sensitive taxa (section 3-4.3, TenBrook et al. 2009a). All plant studies were considered chronic because the typical endpoints of growth or reproduction are inherently chronic. Four studies yielding four plant toxicity values were rated RR for the chronic criterion derivation (Tables 6).

Plant studies are more difficult to interpret than animal data because a variety of endpoints may be used, but the significance of each one is less clear. In this methodology, only endpoints of growth or reproduction (measured by biomass) and tests lasting at least 24-h had the potential to be rated highly and used for criteria calculation, which is in accordance with standard methods (ASTM 2007a, 2007b; USEPA 1996). The plant studies were rated for quality using the data evaluation criteria described in the methodology (section 3-2.2, TenBrook et al. 2009a).

6 Data reduction

Multiple toxicity values for trifluralin for the same species were reduced down to one species mean acute value (SMAV) or one species mean chronic value (SMCV) according to procedures described in the methodology (section 3-2.4, TenBrook et al. 2009a). One study was reduced from the final acute data set (Table 4). The final chronic data set was reduced by one alga, one plant, and one animal value (Table 8).

7 Acute criterion calculation

An acute criterion was calculated with acute animal toxicity data only, because plant toxicity tests are always considered chronic (section 3-2.1.1.1, TenBrook et al. 2009a). Since acceptable acute toxicity values were not available from the five required taxa for a species sensitivity distribution, the acute criterion was calculated using the Assessment Factor (AF) procedure (section 3-3.3, TenBrook et al. 2009a). Trifluralin is an organic pesticide, and the AFs given in the methodology (Table 3.13, TenBrook et al. 2009a) are the most specific AFs available for organic pesticides. The methodology points out that the AFs are limited in that they are based on organochlorine, organophosphate, and pyrethroid pesticides, which are neurotoxic insecticides, while trifluralin is an organofluorine herbicide that inhibits meristem growth. However, trifluralin does exhibit toxicity to animals with an unclear mechanism and is an organic pesticide, thus, it is reasonable to use the AF procedure for trifluralin.

The AFs given in the methodology will be used for trifluralin with the understanding that AFs based on measured pesticide toxicity data are likely more accurate than choosing an arbitrary AF. The methodology points out that AFs are recognized as a conservative approach for dealing with uncertainty in assessing risks posed by chemicals (section 2-3.2, TenBrook et al. 2009a). Using an AF to calculate a criterion always involves a high degree of uncertainty and there is potential for under- or over-protection, which is strongly dependent on the representation of sensitive species in the available data set. The methodology instructs that the derived criterion should be compared to all available ecotoxicity data to ensure that it will be protective of all species (section 3-6.0, TenBrook et al. 2009a).

There are two available taxa in the acceptable (RR) data set shown in the in Table 3: planktonic crustacean (*Daphnia magna*) and warm water fish (*Pimephales promelas* and *Cyprinus carpio*). Missing from the taxa requirements for use of a species sensitivity distribution (SSD) are a Salmonidae, a benthic crustacean, and an insect. The AF method calculates the criterion by dividing the lowest SMAV from the acceptable (RR) data set by an AF, which is determined by the number of taxa available in the data set (section 3-3.3, TenBrook et al. 2009a). The lowest SMAV was the 96-h *Cyprinus carpio* LC₅₀ value of 45 µg/L. This value was divided by an AF of 12 because there are acceptable data from two taxa (Table 17, Fojut et al. 2014). The acute value calculated using the AF represents an estimate of the median 5th percentile value of the SSD, which is the recommended acute value. The recommended acute value is divided by a factor of 2 to calculate the acute criterion (section 3-3.3, TenBrook et al. 2009a). Because the toxicity data used to calculate the criterion only reported two significant figures, the criterion is rounded to two significant figures (section 3-3.2.6, TenBrook et al. 2009a).

```
Acute value = lowest value in data set \div assessment factor = 45 mg/L \div 12 = 3.75 \mug/L 
Acute criterion = acute value \div 2 = 3.75 \mug/L \div 2 = 1.875 \mug/L
```

Acute criterion = $1.9 \mu g/L$

8 Chronic criterion calculation

Although trifluralin is an herbicide, the chronic data in Tables 6 and 7 demonstrate that plants are not the most sensitive taxa; therefore, the procedure for derivation of the chronic criterion of an herbicide was not followed (section 3-4.3, TenBrook et al. 2009a). The chronic criterion was derived using animal data. Acceptable chronic animal values were not available for five different species, so a distribution could not be fit to the available toxicity data (section 3-4.1, TenBrook et al. 2009a). The methodology instructs that in the absence of acceptable data to fit a distribution, the

chronic criterion is calculated using an acute-to-chronic ratio (ACR) (section 3-4.2, TenBrook et al. 2009a). The study parameters for an ACR based on measured data were not met (section 3-4.2.1, TenBrook et al. 2009a). Only one study using *Pimephales promelas* was available that reported acute and a chronic toxicity values, however, the study was performed under static renewal conditions rather than flow-through (Macek et al. 1976). Default ACR values were used to calculate the chronic criterion using animal data (section 3-4.2.4, TenBrook et al. 2009a). The acute 5^{th} percentile value was estimated as $3.75 \,\mu\text{g/L}$ by the assessment factor (AF) method using the LC₅₀ of $45 \,\mu\text{g/L}$ for *Cyprinus carpio* (Poleksic 1999) and an AF of 12 (see Acute Criterion calculation in Section 7). The default ACR of 11.4 (Fojut et al. 2014) was then used to calculate the chronic criterion. Because the toxicity data used to calculate the criterion only reported two significant figures, the criterion is rounded to two significant figures (section 3-3.2.6, TenBrook et al. 2009a).

```
Selected percentile value = estimated 5<sup>th</sup> percentile value = lowest value in data set \div assessment factor = 45 \mu g/L \div 12 = 3.75 \mu g/L
```

```
Chronic criterion = (Selected percentile value) \div ACR
= (3.75 \mu g/L) \div 11.4
= 0.3289
```

Chronic criterion = $0.33 \mu g/L$

9 Water quality effects

9.1 Bioavailability

Few studies were found concerning the bioavailability of trifluralin, and only one study was found pertaining to bioavailability to organisms in the water column. Yockim et al. (1980) found that bioavailability to aquatic organisms was dependent on the rate of trifluralin desorption from the flooded sediment. In a static test there were no toxic effects to daphnids, snails, algae, or mosquito fish due to trifluralin remaining sorbed to the organic matter and clay constituents in the sediment. During a flow-through experiment there were toxic effects to the algae and fish although the concentration of trifluralin in the water was on average three times higher than in the static test. No information about bioavailability of trifluralin in the water column that differentiates between sorption to solids or to dissolved solids or as freely dissolved compound was found. Until there is more information that discusses the bioavailability of these three phases, compliance must be based on the total concentration of trifluralin in water (section 3-5.1, TenBrook et al. 2009a).

9.2 Mixtures

The concentration addition model and the non-additive interaction model are the only predictive mixture models recommended by the methodology (section 3-5.2, TenBrook et al. 2009a), so other models found in the literature will not be considered for compliance. Trifluralin can occur in the environment with other herbicides of similar or different modes of action. Trifluralin is a dinitroaniline herbicide that acts as a meristem growth inhibitor.

Only one study was available that explored toxicity mixture effects of meristem inhibitor herbicides such as trifluralin on aquatic species. George and Liber (2007) studied a mixture of three chemicals with different modes of action with *Daphnia magna*. The mixture was composed of trifluralin and the two insecticides chlorpyrifos and endosulfan. It was found that the mixture toxicity was predicted with a response addition model. This is a noninteractive joint action model for mixtures of compounds with dissimilar modes of action and therefore will not be considered for criteria compliance.

9.3 Temperature, pH, and other water quality effects

Temperature, pH, and other water quality effects on the toxicity of trifluralin were examined to determine if any effects are described well enough in the literature to incorporate into criteria compliance (section 3-5.3, TenBrook et al. 2009a). There were no studies available that examined the effect of pH on toxicity in the aqueous environment. As trifluralin is a weak base, pH is not expected to have a significant effect on the chemical structure in the range of conditions found in natural freshwater environments.

Macek et al. (1969) tested the effect of temperature on fishes in 96 hour static tests. The toxicity of *O. mykiss* to trifluralin was shown to be five times greater when tested at 12.7 °C compared to 1.6 °C. Similarly, *L. macrochirus* was four times more susceptible to trifluralin at 23.8 °C compared to 12.7 °C.

Until more data becomes available for relevant species, it is not possible to quantify the relationship between the toxicity of trifluralin and temperature for water quality criteria at this time (section 3-5.3, TenBrook et al. 2009a).

10 Comparison of ecotoxicity data to derived criteria

10.1 Sensitive species

The derived criteria were compared to toxicity values for the most sensitive species in both the acceptable (RR) and supplemental (RL, LR, LL) data sets to ensure that these species will be adequately protected (section 3-6.1, TenBrook et al. 2009a).

The lowest acute value in the data sets rated RR, RL, LR, or LL (Tables 3, 4, and 5) is 41 µg/L for rainbow trout, *O. mykiss* (Johnson & Finley 1980). This study rated LL because a standard method was not followed and the control response was not reported. In addition, the reliability score was low due to a lack of parameter reporting. These factors make this study less reliable for the purposes of the methodology, but it is still a relevant toxicity study. This study tested an aquatic species that resides in North America with the endpoint and exposure duration fit into the acute test definition in the methodology (section 3-2.1.1.1). The next lowest acute value is 45 µg/L for carp (*Cyprinus carpio*, Poleksic 1999), which is rated RR. The derived acute criterion (1.9 µg/L) is also based on this species and appears to be protective of all sensitive species int he data sets.

The chronic animal data set shows that aquatic animals are more sensitive to trifluralin than plants. The chronic criterion was therefore calculated to be protective of animals (0.33 μ g/L) and is an order of magnitude lower than the lowest chronic plant MATC of 7.9 μ g/L for *Raphidocelis subcapitata* and the lowest chronic animal MATC of 3.1 for *Pimephales promelas*. Adequate protection will be attained for these sensitive species.

10.2 Ecosystem and other studies

The derived criteria are compared to acceptable laboratory, field, or semi-field multispecies studies (rated R or L) to determine if the criteria will be protective of ecosystems (section 3-6.2, TenBrook et al. 2009a). No acceptable mesocosm, microcosm or ecosystem (field and laboratory) studies were identified. One microcosm study was available that rated N and another was located that used a formulated product.

10.3 Threatened and endangered species

The derived criteria are compared to measured toxicity values for threatened and endangered species (TES), as well as to predicted toxicity values for TES, to ensure that they will be protective of these species (section 3-6.3, TenBrook et al. 2009a). Current lists of state and federally listed threatened and endangered plant and animal species in California were obtained from the California Department of Fish and Game website (CDFG 2015). One listed animal species is represented in the dataset. Five Evolutionarily Significant Units of *Oncorhynchus mykiss* are listed as federally threatened or endangered throughout California. A supplemental acute study that rated LL due to lack of a standard method and control results reported a LC₅₀ of 41 μ g/L for *O. mykiss* (Johnson & Finley 1980). This data indicates that the acute criterion of 1.9 μ g/L would be protective of this species.

The USEPA interspecies correlation estimation (ICE v. 3.1; USEPA 2010) software was used to estimate toxicity values for the listed animals or plants represented in the acute data set by members of the same family or genus. Table 11 summarizes the results of the ICE analyses. The estimated toxicity values in Table 11 range from 63.67 $\mu g/L$ for Chinook salmon, 53.70 $\mu g/L$ Coho salmon, 25.31 $\mu g/L$ for Apache trout, and 48.61 $\mu g/L$ for Cutthroat salmon.

No plant studies used in the criteria derivation were of state or federal endangered, threatened or rare species. Plants may be particularly sensitive to trifluralin because it is an herbicide, but there are no aquatic plants listed as state or federal endangered, threatened or rare species so they could not be considered in this section.

Based on the available data and estimated values for animals, there is no evidence that the value referenced in place of a calculated acute and or the calculated chronic criteria will be underprotective of threatened and endangered species.

11 Harmonization with other environmental media

11.1 Bioaccumulation

Bioaccumulation was assessed to ensure that the derived criteria will not lead to unacceptable levels of trifluralin in food items (section 3-7.1, TenBrook et al. 2009a). Trifluralin has a log K_{ow} of 4.64 (Section 3), a K_d of 105-1217 depending on soil type (Cooke et al. 2000; Ying and Williams 2000), and a molecular weight of 335.29, which indicates bioaccumulative potential. There are no FDA action levels for trifluralin in food (USFDA 2000), and there are no EPA pesticide tolerances set for any aquatic species (USEPA 2013). Bioconcentration of trifluralin has been measured in unknown species (Table 1).

To check that these criteria are protective of terrestrial wildlife that may consume aquatic organisms, a bioaccumulation factor (BAF) was used to estimate the water concentration that would roughly equate to a reported toxicity value for such terrestrial wildlife (LC_{50, oral predator}). These calculations are further described in section 3-7.1 of the methodology (TenBrook et al. 2009a). The BAF of a given chemical is the product of the BCF and a biomagnification factor (BMF), such that BAF=BCF*BMF. No BMF value was found for trifluralin. Chronic dietary toxicity values are preferred for this calculation. The BAF and BCF values available were either from an estimation modeling program (USEPA 2015) or the value origin was not reported (PPDB 2015). A single dietary value for mallard was determined to be 910.50 mg/kg from a highly rated study (Beavers et al. 1987). A BCF of 2363.38 L/kg (USEPA 2015 and PPDB 2015) were used as an example estimation of bioaccumulation in the environment. No BMF value was available in the literature so it was estimated two ways according to the methodology (a value of 2 both when as approximated from log K_{ow} and as approximated from BCF as in section 3-7.1 and Table 3.15 in TenBrook et al. 2009a).

$$NOEC_{water} = \frac{NOEC_{oral-predator}}{BCF_{food_item} \cdot BMF_{food_item}}$$

Mallard:
$$NOEC_{water} = \frac{910.50^{mg}/kg}{1905.11^{L}/kg} * 2 = 0.2390^{mg}/L = 239.0^{\mu g}/L$$

In this example, the calculated chronic criterion (0.33 μ g/L) is more than two orders of magnitude below the estimated NOEC_{water} value for wildlife and is not expected to cause adverse effects due to bioaccumulation.

11.2 Harmonization with air and sediment criteria

This section addresses how the maximum allowable concentration of trifluralin might impact life in other environmental compartments through partitioning (section 3-7.2, TenBrook et al. 2009a). One sediment study was available with a EC₅₀ value of 6,600 µg/L and a NOEC value of 250 µg/L for the sediment-dwelling larvae of *Chironomus riparius* (Knoch 1996). The derived criteria are well below the effect levels for *C. riparius*. The other available sediment value for trifluralin is estimated based on partitioning from water using empirical K_{oc} values. These range from 4.215 µg/L to 4.252 µg/L (USEPA 2015). Trifluralin is listed as a hazardous air pollutant and toxic air contaminant by the California Air Resources Board (CCR 2016) although a reference concentration for chronic inhalation exposure is not available (IRIS 1989). There are no other federal or state sediment or air quality standards for trifluralin (CARB 2008; CDWR 1995), nor is trifluralin mentioned in the NOAA sediment quality guidelines (NOAA 1999). For biota, the limited data on bioconcentration or biomagnification of trifluralin is addressed in section 15.

12 Trifluralin criteria summary

12.1 Limitations, assumptions, and uncertainties

The assumptions, limitations and uncertainties involved in criteria generation are available to inform environmental managers of the accuracy and confidence in criteria (section 3-8.0, TenBrook et al. 2009a). Chapter 2 of the methodology (TenBrook et al. 2009a) discusses these points for each section as different procedures were chosen, such as the list of assumptions associated with using an SSD (section 2-3.1.5.1), and reviews them in section 2-7.0. This section summarizes any data limitations that affected the procedure used to determine the final trifluralin criteria.

Overall, there was a lack a highly rated aquatic plant and animal toxicity data for trifluralin. Both the acute and chronic data sets lacked the full complement of five required taxa to fit a distribution for criteria derivation. The acute data set was missing values for a Salmonidae, a benthic crustacean, and an insect. The AF procedure was used to calculate the acute criterion.

The chronic data set contained only four out of five different species of vascular plants or alga, however for trifluralin, animals were more sensitive than plants and alga based on the available data. Chronic animal taxa requirements were not met, as there

were only three values available. The chronic criterion was derived with a minimum amount of data according to the methodology (section 3-4.2.3, TenBrook et al. 2009a) using chronic animal data and a default ACR.

Other limitations include the lack of sediment, bioavailability, and wildlife studies. Only one study sediment study was available and was not sufficient to assess partitioning of trifluralin from other environmental niches than the water column. One bioavailability study was available for organisms in the water column. Additional high quality mallard duck studies are needed to determine definitive toxicity values.

12.2 Comparison to national standard methods

This section is provided as a comparison between the UC-Davis methodology for criteria calculation (TenBrook et al. 2009a) and the current USEPA (1985) national standard. The following example trifluralin criteria were generated using the USEPA (1985) methodology with the data set generated in this trifluralin criteria report.

The USEPA acute methods have three additional taxa requirements beyond the five required by the SSD procedure of the UC-Davis methodology (section 3-3.1, TenBrook et al. 2009a). They are:

- 1. A third family in the phylum Chordata (e.g., fish, amphibian);
- 2. A family in a phylum other than Arthropoda or Chordata (e.g., Rotifera, Annelida, Mollusca):
- 3. A family in any order of insect or any phylum not already represented.

None of the three additional requirements could be met. Missing from the USEPA (1985) methodology requirements are a warm water fish, a benthic crustacean, an insect, a third family in the phylum Chordata, and a family in a phylum other than Arthropoda or Chordata. Because of this lack of data, no acute criterion could be calculated according to the USEPA (1985) methodology.

According to the USEPA (1985) methodology, the chronic criterion is equal to the lowest of the Final Chronic Value, the Final Plant Value, and the Final Residue Value.

To calculate the Final Chronic Value, animal data is used and the same taxa requirements must be met as in the calculation of the acute criterion (section III B USEPA 1985). Three of the eight taxa requirements are available in the RR chronic animal data set with *Daphnia magna*, *Oncorhynchus mykiss*, and *Pimephales promelas*. (Table 7). The missing taxa are as follows:

- 1. A benthic crustacean
- 2. An insect (aquatic exposure)
- 3. A third family in the phylum Chordata (e.g., fish, amphibian)
- 4. A family in a phylum other than Arthropoda or Chordata (e.g., Rotifera, Annelida, Mollusca)
- 5. A family in any order of insect or any phylum not already represented

The California Department of Fish and Game has derived criteria using the USEPA (1985) SSD method with fewer than the eight required families, using professional judgment to determine that species in the missing categories were relatively insensitive and their addition would not lower the criteria (Menconi & Beckman 1996; Siepmann & Jones 1998). In this case, there are too many missing taxa values to derive a Final Chronic Value in this way.

The Final Plant Value is calculated as the lowest result from a 96-hr test conducted with an important plant species in which the concentrations of test material were measured and the endpoint was biologically important. None of the plant toxicity values in the RR data set (Table 6) are for a 96-hr test; they are longer ranging from five to 14 days. The lowest NOEC reported is $5.37~\mu g/L$ for *Raphidocelis subcapitata* (Adams 1990a) to serve as the chronic criterion. This test has an exposure duration that is one day longer than the specified duration.

```
Final Plant Value = lowest result from a plant test
= 5.37 \mu g/L
```

The Final Residue Value is calculated by dividing the maximum permissible tissue concentration by an appropriate bioconcentration or bioaccumulation factor. A maximum allowable tissue concentration is either (a) a FDA action level for fish oil or for the edible portion of fish or shellfish, or (b) a maximum acceptable dietary intake based on observations on survival, growth, or reproduction in a chronic wildlife feeding study or long-term wildlife field study. There are no FDA action levels for trifluralin in food (USFDA 2000) and there are no EPA pesticide tolerances set for any aquatic species (USEPA 2013). A single dietary NOEC of 910.50 mg/kg (Beavers et al. 1987) was the lowest wildlife dietary toxicity value available. A BCF of 1905.11 for an unknown species (Table 1) is used to calculate the Final Residue Value.

```
Final Residue Value = maximum acceptable dietary intake \div BCF = 910.50 mg/kg \div 1905.11 L/kg = 0.478 mg/L = 4,780 \mug/L
```

The Final Plant Value is lower than the Final Residue Value. A Final Chronic Value cannot be calculated. Therefore the chronic criterion by the USEPA (1985) methodology for trifluralin would be $5.37~\mu g/L$. The example chronic criterion is higher than the one recommended by the UC-Davis methodology.

12.3 Final criteria statement

The final criteria statement is:

Aquatic life in the Sacramento River and San Joaquin River basins should not be affected unacceptably if the four-day average concentration of trifluralin does not exceed

 $0.33 \mu g/L$ more than once every three years on the average and if the one-hour average concentration does not exceed 1.9 $\mu g/L$ more than once every three years on the average.

Although the criteria were derived to be protective of aquatic life in the Sacramento and San Joaquin Rivers, these criteria would be appropriate for any freshwater ecosystem in North America, unless species more sensitive than are represented by the species examined in the development of these criteria are likely to occur in those ecosystems.

The acute criterion is based only on acute animal data and was derived to protect animals from acute pulses of trifluralin. Details of the acute criterion calculation are described in section 7 and the acute data are shown in Tables 3 - 5. An assessment factor was used instead of a distribution to calculate the acute criterion because there were not sufficient data from the five required taxa for use of a SSD.

Details of the chronic criterion calculation are described in section 8 and chronic plant data are shown in Table 6 Although trifluralin is an herbicide it was shown that aquatic animals display a higher sensitivity. The chronic criterion was calculated using animal data by the ACR method because there was insufficient data for use of a SSD for criterion calculation.

There are no established water quality criteria for trifluralin with which to compare the criteria derived in this report. The US EPA has several aquatic life benchmarks established for trifluralin, shown in Table 11, to which the derived criteria in this report can be compared with caution (USEPA 2014). According to the USEPA (2014), aquatic life benchmarks are not calculated following the same methodology used to calculate water quality criteria. Water quality criteria can be used to set water quality standards under the Clean Water Act, but aquatic life benchmarks may not be used for this purpose (USEPA 2014).

The referenced acute value in this report is well below both the acute fish benchmark and the acute invertebrate benchmark by factors of more than 32 and 400 times, respectively (Table 12). The derived chronic criterion of this report is well below the chronic benchmarks for fish and invertebrates as well as acute nonvascular plants (by factors of 2.5, 5, and 16, respectively). Although trifluralin is an herbicide, aquatic animals exhibit a higher sensitivity than plants. This is reflected in the US EPA benchmarks.

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Data Tables

Table 3 Final acute toxicity data set for trifluralin.

All studies were rated RR and were conducted at standard temperature. S: static; SR: static renewal; FT: flow-through.

Species	Common Identifier	Family	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/ size	$\begin{array}{c} LC/EC_{50}\left(\mu g/L\right)\\ (95\%\ CI) \end{array}$	Reference
Cyprinus									6 mo/3.3 cm, 0.39		Poleksic
carpio	Carp	Cyprinidae	SR	NR	99.00%	96 h	20	Mortality	g	45 (36-51)	1999
Daphnia magna	Daphnid	Daphniidae	S	Meas	97.10%	48 h	20	Mortality	<24 h	245 (130-438)	Kirk 1999
Pimephales promelas	Fathead minnow	Cyprinidae	SR	Meas	97.00%	48 h	25	Mortality	26 d	115 (48-211)	Macek 1976

Table 4 Acceptable reduced acute data rated RR. Reduction reason given. S: static; SR: static renewal; FT: flow-through.

										LC/EC ₅₀		
Species	Common Identifier	Family	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/size	(μg/L) (95% CI)	Reference	Reason
										193		
Daphnia										(115-	Macek	
magna	Daphnid	Daphniidae	S	Nom	97.00%	48 h	20	Mortality	<24 h	327)	1976	С

Reduction Reasons

A. Data calculated from nominal concentrations

Table 5 Supplemental acute data rated RL, LR, LL. Exclusion reasons given. S: static; SR: static renewal; FT: flow-through. NR: not reported. 95% CI: 95% confidence interval. Exclusion reasons are listed at the end of the table.

Species	Common Identifier	Family	Test type	Meas / Nom	Chemica l grade	Duration	Tem p (°C)	Endpoint	Age/ size	LC/EC ₅ ₀ (μg/L) (95% CI)	Referenc e	Rating/ Reason
Anazyrus fowleri	Western chorus frog	Bufonidae	S	Nom	Technical	96 h	15.5	Mortality	Tadpole s	100 (80- 490)	Sanders 1970	1, 3, 4
Carrassius auratus	Goldfish	Cyprinidae	S	Nom	95.90%	96 h	18	Immobilizatio n	1.0 g	145 (108- 195)	Johnson 1980	1, 3, 4
Cyprinodon variegatus	Sheepshead minnow	Cyprinodontida e	SR	Meas	99.00%	96 h	30	Mortality	Fry, 1 cm	190 (128- 282)	Parrish 1978	2
Lepomis macrochirus	Bluegill sunfish	Centrarchidae	S	Nom	95.90%	96 h	22	Immobilizatio n	0.8 g	58 (47- 70)	Johnson 1980	1, 3, 4
Micropterus salmoides	Largemouth bass	Centrarchidae	S	Nom	95.90%	96 h	18	Immobilizatio n	0.7 g	75 (65- 87)	Johnson 1980	1, 3, 4
Onchorhynchus mykiss	Rainbow trout	Salmonidae	S	Nom	95.90%	96 h	12	Immobilizatio n	0.8	41 (26- 62)	Johnson 1980	1, 3, 4
Pimephales promelas	Fathead minnow	Cyprinidae	S	Nom	95.90%	96 h	18	Immobilizatio n	0.8	105 (83- 134)	Johnson 1980	1, 3, 4

Exclusion Reasons

- 1. Not a standard method
- 2. Saltwater
- 3. Control response low or not reported
- 4. Low reliability score

Table 6 Final chronic plant toxicity data set for trifluralin.
All studies were rated RR. S: static; SR: static renewal; FT: flow-through. NR: not reported, n/a: not applicable.

Species	Common identifier, Family	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/size	NOEC (μg/L)	LOEC (µg/L)	MATC (μg/L)	EC ₅₀ (μg/L)	Reference
Anabena flos-aquae	Cyanobacterium	S	Meas	97.92%	5 d	24	Cell count	Algal cells	339	NR	NR	>2S	Hughes 1993a
Lemna gibba	Duckweed	S	Meas	Technical	14 d	25	Growth	1 w	12.9	25.3	18	55.9 (38.96- 72.74)	Milazzo 1993
Navicula pelliculosa	Diatom	S	Meas	97.92%	5 d	24	Cell count	Algal cells	<7.65	NR	NR	15.3 (6.72- 34.7)	Hughes 1993b
Raphidocelis subcapitata	Alga	S	Meas	99.86%	7 d	22	Cell count	Algal cells	5.37	11.7	7.9	12.2	Adams 1990a

Table 7 Final chronic animal toxicity data set for trifluralin.
All studies were rated RR. S: static; SR: static renewal; FT: flow-through. NR: not reported

Species	Common identifier	Test type	Chemical grade	Duration	Age/size	NOEC (μg/L)	LOEC (µg/L)	MATC (μg/L)	Reference
Daphnia magna	Daphnid	S	97.00%	64 d	<24 h	2.4	7.2	4.2	Macek 1976
Oncorhynchus mykiss	Rainbow trout	FT	83.30%	48 d	Eye stage eggs	1.14	8.81	3.17	Adams 1990b
Pimephales promelas	Fathead minnow	SR	97.00%	61 w	26 d	1.9	5.1	3.1	Macek 1976

Table 8 Acceptable reduced chronic data rated RR. Exclusion reason given. S: static; SR: static renewal; FT: flow-through. NR: not reported

Species	Common identifier	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/size	NOEC (µg/L)	LOEC (µg/L)	MATC (μg/L)	Reference	Reason for exclusion
Raphidocelis subcapitata	Alga	S	Nom	Technical	96 h	25	Biomass	Algal cells	150	300	212.1	Fairchild 1997	A
Lemna gibba	Duckweed	S	Nom	Technical	96 h	25	Biomass	NR	75	150	106	Fairchild 1997	A
Daphnia magna	Daphnid	SR	Meas	99.86%	21 d	20	Mortality	<24 h	50.7	NR	NR	Grothe 1990	В

Exclusion Reasons

A. Data calculated from nominal concentrations

B. Less sensitive time point

Table 9 Supplemental chronic plant toxicity data set of studies rated RL, LR, or LL.

S: static; SR: static renewal; FT: flow-through. NR: not reported, n/a: not applicable; 95% CI: 95% confidence interval; SE: standard error.

Species	Common identifier	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/ size	NOEC (µg/L)	LOEC (µg/L)	EC ₅₀ (μg/L) (95% CI)	Reference	Rating/ Reason for exclusion
<u> </u>											28		
Skeletonema								Algal			(24.2-		
costatum	Diatom	S	Meas	97.92%	5 d	20	Cell count	cells	4.6	NR	32.5)	Hughes 1993c	1

Exclusion Reasons

1. Not a standard method

Table 10 Supplemental chronic animal toxicity data for studies rated RL, LR, or LL.

S: static; SR: static renewal; FT: flow-through. NR: not reported; 95% CI: 95% confidence interval.

Species	Common identifier	Test type	Meas /Nom	Chemical grade	Duration	Tem p (°C)	Endpoint	Age/siz e	NOE C (µg/L)	LOEC (µg/L)	MATC (μg/L) (95% CI)	Reference	Rating/ Reason for exclusion
Cyprinodon	Sheepshead											Parrish	
variegatus	minnow	SR	Meas	99.00%	166 d	30	Mortality	Fry	1.3	4.8	2.50	1978	1

Exclusion Reasons

1. Saltwater

Table 11 US EPA Aquatic Life Benchmarks.				
All units are μg/L. (USEPA 2009)				
Acute Fish	Chronic Fish	Acute	Chronic	Acute
		Invertebrates	Invertebrates	nonvascular
				plants
20.5	1.14	280	2.4	7.52

Appendix A - Aqueous Toxicity Data Summaries

Appendix A1 - Aqueous Toxicity Studies Rated RR

Anabena flos-aquae

Study: Hughes JS and Williams TL. (1993a) The toxicity of trifluralin to *Anabena flos-aquue*. Malcolm Pirnie, Inc., Tarrytown, New York. Laboratory study number B460-153-1. Submitted to The Dow Chemical Company, Indianapolis, Indiana. USEPA MRID 42834103.

RelevanceReliabilityScore: 100Score: 94.5Rating: RRating: R

	Hughes & Williams 1993a	A. flos-aquae
Parameter	Value	Comment
Test method cited	Pesticide Assessment	
	Guidelines, USEPA	
Phylum/subphylum	Cyanobacteria	
Order	Nostocales	
Family	Nostocaceae	
Genus	Anabena	
Species	Flos-aquae (Lyng.) Breb.	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Algal cells	
Source of organisms	Laboratory cultures	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	Given organism size and presence in growth medium, it is assumed that aliquots are inherently randomly
Test vessels randomized?	Not reported	
Test duration	5 d	
Data for multiple times?	3, 4, 5 d	
Effect 1	Cell count	
Control response 1	351,000	
Temperature	24 ± 2 °C	
Test type	Static	
Photoperiod/light intensity	Continuous/200 footcandles	
Dilution water	Synthetic AAP growth	Made with Type I
	medium	water
Feeding	Growth medium	
Purity of test substance	97.92 %	
Concentrations measured?	Yes	

	Hughes & Williams 1993a	A. flos-aquae
Parameter	Value	Comment
Measured is what % of nominal?	105-129 %	
Toxicity values calculated based on	Initial measured	
nominal or measured		
concentrations?		
Chemical method documented?	HPLC	
Concentration of carrier (if any) in	Dimethylformamide, 0.48	
test solutions	mL/L	
Concentration 1 Nom; Meas (µg/L)	9.70; 12.5	3 reps, 3,000
		cells/rep
Concentration 2 Nom; Meas (µg/L)	19.3; 21.6	3 reps, 3,000
, , ,		cells/rep
Concentration 3 Nom; Meas (µg/L)	38.6; 45.7	3 reps, 3,000
		cells/rep
Concentration 4 Nom; Meas (µg/L)	77.3; 89.3	3 reps, 3,000
		cells/rep
Concentration 5 Nom; Meas (µg/L)	154; 162	3 reps, 3,000
		cells/rep
Concentration 6 Nom; Meas (µg/L)	308; 339	3 reps, 3,000
		cells/rep
Control	Negative: 0; 0	3 reps, 3,000
	Solvent: 0; 0	cells/rep
EC ₅₀ (95% CI) (μg/L)	>2S	Method: Weighted
		least squares
		nonlinear regression
NOEC	339	Method:
		p:
		MSD:
LOEC	Not reported	
MATC	Not reported	
% control at NOEC	80 %	280,667 (tmt) /
		351,000 (mean
		controls) = 80
% control at LOEC	Not calculable	

Notes: Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires Type I water, and the medium is presumably appropriate for the test species because a specific culture media was used. Trifluralin solubility (S) = $238.08 \, \mu g/L$, $2S = 476.16 \, \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Statistical significance (2), Minimum significant difference (2). Total: 100-4=96

<u>Acceptability:</u> Temperature variation (3), Random design (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-7 = 93

Reliability score: mean(96,93)=94.5

Cyprinus carpio

Study: Poleksić, V. and Karan, V., 1999. Effects of trifluralin on carp: biochemical and histological evaluation. *Ecotoxicology and Environmental Safety*, 43(2), pp.213-221.

RelevanceReliabilityScore: 92.5Score: 75.5Rating: RRating: R

Relevance points taken off for: Control response (7.5). 100-7.5=92.5

	Poleksic & Karan 1999	C. carpio
Parameter	Value	Comment
Test method cited	OECD Guidelines, Numbers	
	203-204, 1984, 1987, 1990	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Cyprinus	
Species	Carpio	
Family native to North America?	Introduced	
Age/size at start of test/growth	Acute: 6 month old, 3.3 cm,	
phase	0.39 g	
	Subacute: 1.5 year, 15.3 cm,	
	154.6 g	
Source of organisms	Ecka fish farm	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	21 d	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	Acute: 96 h	
	Subacute: 14 d	
Data for multiple times?	Acute: 24, 48,96 h	
	Subacute:	
Effect 1	Mortality	
Control response 1	Subacute: 100% survival	
Effect 2	Body weight	
Control response 2	Subacute: Not reported	
Temperature	20 ± 1 °C	
Test type	Static renewal	
Photoperiod/light intensity	12:12, 1:d	
Dilution water	Dechlorinated tap water	
рН	7.8-8.2	

	Poleksic & Karan 1999	C. carpio
Parameter	Value	Comment
Hardness	150-230 mg/L CaCO ₃	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	
Feeding	Aquaria fish mixture	Once daily
Purity of test substance	99 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	Not reported	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Acute: 25; Not reported	Acute: 0 reps,
	Subacute: 5; Not reported	10/rep
		Subacute: 0 reps,
		8/rep
Concentration 2 Nom; Meas (µg/L)	Acute: 50; Not reported	
, ,	Subacute: 10; Not reported	
Concentration 3 Nom; Meas (µg/L)	Acute: 100; Not reported	
, ,	Subacute: 20; Not reported	
Concentration 4 Nom; Meas (µg/L)	Acute: 200; Not reported	
Control	Acute: 0; Not reported	
	Subacute: 0; Not reported	
LC ₅₀ (95% CI) (µg/L)	Acute:	Method: Litchfield
,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	24 h: 185 (173-189)	and Wilcoxon
	48 h: 66 (54-75)	
	96 h: 45 (36-51)	

Notes: Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires deionized water, and the medium is presumably appropriate for the test species because a specific culture media was used.

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Measured concentrations (3), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), Hypothesis tests (8). Total: 100- 19=81

<u>Acceptability:</u> Control response (9), Measured concentrations within 20% nominal (4), Alkalinity (2), Dissolved oxygen (6), Conductivity (1), Number of concentrations (3), Random design (2), Hypothesis tests (3). Total: 100-30 = 70

Reliability score: mean(81,70)=75.5

Daphnia magna

Study: Grothe DW and Mohr RR. (1990) The chronic toxicity of trifluralin to *Daphnia magna* in a static renewal life-cycle test. Lilly Research Laboratories, Greenfield, Indiana. Laboratory project identification C01589. USEPA MRID 41386201.

RelevanceReliabilityScore: 100Score: 88.5Rating: RRating: R

	Grothe & Mohr 1990	D. magna
Parameter	Value	Comment
Test method cited	OECD, 1984; USEPA,	
	1987	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	First instar, <24 h	
phase		
Source of organisms	Laboratory cultures	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Yes	
Test duration	21 d	
Data for multiple times?	No	
Effect 1	Survival	
Control response 1	100 %	
Effect 2	Days to brood	
Control response 2	9.5 d	
Effect 3	Neonates/female	
Control response 3	112.4 %	
Effect 4	Body length	
Control response 4	4.42 mm	
Temperature	20.3 ± 0.4 °C	
Test type	Static renewal	
Photoperiod/light intensity	16:8, 1:d/<1 footcandle	
Dilution water	Well water	
pH	7.7-8.6	
Hardness	120-137 mg/L CaCO ₃	

	Grothe & Mohr 1990	D. magna
Parameter	Value	Comment
Alkalinity	145-150 mg/L CaCO ₃	
Conductivity	232-291 μS/cm	
Dissolved Oxygen	7.6 mg/L	84 %
Feeding	Green algal cells and	S. capricornutum
	cerophyl	
Purity of test substance	99.86 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	70-76%	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	2.25; 1.57	10 reps, 1/rep
Concentration 2 Nom; Meas (µg/L)	4.5; 3.19	
Concentration 3 Nom; Meas (µg/L)	9; 6.53	
Concentration 4 Nom; Meas (µg/L)	18.0; 13.7	
Concentration 5 Nom; Meas (µg/L)	36.0; 26.2	
Concentration 6 Nom; Meas (µg/L)	72.0; 50.7	
Control	Negative: 0; 0	
	Solvent: 0; 0	
NOEC	50.7	Method: n/a
		p: NR
		MSD: NR
% control at NOEC	Survival: 80 %	Survival:
	Days to brood: 95 %	80 (tmt) / 100 (mean
	Neonates/female: 119 %	controls) = 80
	Length: 101 %	Days to brood: 9
		(tmt) / 9.5 (mean
		controls) = 95
		Neonates/female: 134
		(tmt) / 112.5 (mean
		controls) = 119
		Length: 4.48 (tmt) / 4.42 (mean controls)
		= 101
		<u> </u>

Notes: No significant effects seen so NOEC stated as highest exposure concentration.

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Statistical significance (2), Significance level (2), Minimum significant difference (2), Point estimates (8). Total: 100-14 = 86

Acceptability: Carrier solvent (4), Minimum significant difference (1), % control at LOEC (1), Point estimates (3). Total: 100-9 =91

Reliability score: mean(86,91)=88.5

Daphnia magna

Study: Kirk HD, Marino TA, Hugo JM. (1999) Evaluation of the acute toxicity of trifluralin technical expose daphnia, *Daphnia magna* Straus. Health & Environmental Research Laboratories, Midland, Michigan. Study ID 981190R. Dow AgroSciences, LLC, Indianapolis, Indiana. USEPA MRID 4787007.

RelevanceReliabilityScore: 100Score: 91Rating: RRating: R

	Kirk et al. 1999	D. magna
Parameter	Value	Comment
Test method cited	OECD method 202 part I	
	and EC Directives 91/414	
	and 92/69	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	<24 h	
phase		
Source of organisms	Laboratory cultures	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	6, 24, 48 h	
Effect 1	Mortality	
Control response 1	48 h: 0 %	
Temperature	$20 \pm 1 {}^{\circ}\text{C}$	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Lake Huron surface water	Irradiated, filtered in laboratory
pН	7.5-7.9	•
Hardness	170 mg/L CaCO ₃	
Alkalinity	28 mg/L CaCO ₃	
Conductivity	397 μS/cm	
Dissolved Oxygen	8.0-8.8 mg/L	88-97 %

	Kirk et al. 1999	D. magna
Parameter	Value	Comment
Feeding	Not reported	
Purity of test substance	97.1 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	88-121 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC/ECD	
Concentration of carrier (if any) in test solutions	Dimethylformamide	
Concentration 1 Nom; Meas (µg/L)	15.7; 16.7	2 reps, 10/rep
Concentration 2 Nom; Meas (µg/L)	31.3; 38	2 reps, 10/rep
Concentration 3 Nom; Meas (µg/L)	62.5; 67.7	2 reps, 10/rep
Concentration 4 Nom; Meas (µg/L)	125; 130	2 reps, 10/rep
Concentration 5 Nom; Meas (µg/L)	250; 239	2 reps, 10/rep
Concentration 6 Nom; Meas (µg/L)	500; 438	2 reps, 10/rep
Control	Negative: 0; 0	2 reps, 10/rep
	Solvent: 0; 0	
EC ₅₀ (95% CI) (μg/L)	6, 24 h: >438	Method: Binomial
	48 h: 245 (130-438)	probability/non-
		linear interpolation
NOEC	130	Method: Not
		reported
		p: Not reported
1070		MSD: Not reported
LOEC	Not reported	
MATC	Not reported	
% control at NOEC	100 % survival	

Notes: Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Photoperiod (3), Statistical significance (2), Significance level (2), Minimum significant difference (2). Total: 100- 9=91

<u>Acceptability:</u> Organisms randomized (1), Photoperiod (2), Random design (2), Adequate replication (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-9 =91 **Reliability score: mean(91,91)=91**

Daphnia magna

Study: Macek KJ, Lindberg MA, Sauter S, Buxton KS and Costa PA. (1976) Toxicity of Four Pesticides to Water Fleas and Fathead Minnows: Acute and Chronic Toxicity of Acrolein, Heptachlor, Endosulfan, and Trifluralin to the Water Flea (*Daphnia magna*) and the Fathead Minnow (*Pimephales promelas*). EG & G Bionomics, Wareham, Massachusetts. Laboratory contract number 68-001-0738. Submitted to Environmental Research Laboratory Office of Research and Development, US Environmental Protection Agency, Duluth, Minnesota. EPA contract number EPA-600/3-76-099. USEPA MRID 5008271.

RelevanceReliabilityScore: 100Score: 91Rating: RRating: R

	Macek et al. 1976	D. magna
Parameter	Value	Comment
Test method cited	Standard methods for the	
	estimation of water and	
	wastewater (APHA 1971)	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	<24 h	
phase		
Source of organisms	University of New	
	Hampshire, Durham, New	
	Hampshire	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	22 , 43, 64 d	3 generations
Effect 1	Mortality	
Control response 1	Generation 1 (22 d): 85 %	
	Generation 2 (43 d): 80 %	
	Generation 3 (64 d): 75 %	
Effect 2	Reproduction	
Control response 2	Generation 1 (22 d): 29	
	Generation 2 (43 d): 13	
	Generation 3 (64 d): 9	

	Macek et al. 1976	D. magna
Parameter	Value	Comment
Temperature	20 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Continuous/intensity not	
	reported	
Dilution water	Well water	
рН	6.8-7.2	
Hardness	37 mg/L CaCO ₃	
Alkalinity	35 mg/L CaCO ₃	
Dissolved Oxygen	4.6-8.7 mg/L	50-96 %
Feeding	Trout starter	
Purity of test substance	97 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	40-60 %	
Toxicity values calculated based on	Nominal	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acute: Acetone, 43 mg/L	
test solutions	Chronic: none	4 5/
Concentration 1 Nom; Meas (µg/L)	6; 2.4	4 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	12; 7.2	4 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	25; 14	4 reps, 5/rep
Concentration 4 Nom; Meas (µg/L)	50; 25.6	4 reps, 5/rep
Concentration 5 Nom; Meas (µg/L)	100; 52.7	4 reps, 5/rep
Control	0; 0	4 reps, 5/rep
LC ₅₀ (95% CI) (μg/L)	193 (115-327)	Method:
NOEC	2.4 μg/L	Method:
		p:
		MSD:
LOEC	7.2 μg/L	
MATC (GeoMean NOEC, LOEC)	4.2	
% control at NOEC	Survival:	Survival:
	22 d (gen 1): 106 %	22 d (gen 1): 90
	43 d (gen 2): 62.5 %	(tmt) / 85 (control)
	64 d (gen 3): 100 %	= 106
		43 d (gen 2): 50
	Reproduction:	(tmt) / 80 (control)
	22 d (gen 1): 103 %	= 62.5
	43 d (gen 2): 62 %	64 d (gen 3): 75
	64 d (gen 3): 56 %	(tmt) / 75 (control) = 100
		Reproduction:
		22 d (gen 1): 30
		(tmt) / 29 (control)

	Macek et al. 1976	D. magna
Parameter	Value	Comment
		= 103
		43 d (gen 2): 8 (tmt)
		/ 13 (control) = 62
		64 d (gen 3): 5 (tmt)
		/ 9 (control) = 56
% control at LOEC	Survival:	Survival:
	22 d (gen 1): 51 %	22 d (gen 1): 43
	43 d (gen 2): 25 %	(tmt) / 85 (control)
	64 d (gen 3): 0 %	= 51
		43 d (gen 2): 20
	Reproduction:	(tmt) / 80 (control)
	22 d (gen 1): 86 %	= 25
	43 d (gen 2): 115 %	64 d (gen 3): 0 (tmt)
	64 d (gen 3): 0 %	/ 75 (control) = 0
		Reproduction:
		22 d (gen 1): 25
		(tmt) / 29 (control)
		= 86
		43 d (gen 2): 15
		(tmt) / 13 (control)
		64 d (gen 3): 0 (tmt)
		/9 (control) = 0

Notes:

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Conductivity (2), Minimum significant difference (2). Total: 100-4 = 96

<u>Acceptability:</u> Measured concentrations within 20% nominal (4), Carrier solvent (4), Conductivity (1), Random design (2), Adequate replication (2), Minimum significant difference (1). Total: 100-14 =86

Reliability score: mean(96,86)=91

Daphnia magna

Study: Macek KJ, Lindberg MA, Sauter S, Buxton KS and Costa PA. (1976) Toxicity of Four Pesticides to Water Fleas and Fathead Minnows: Acute and Chronic Toxicity of Acrolein, Heptachlor, Endosulfan, and Trifluralin to the Water Flea (*Daphnia magna*) and the Fathead Minnow (*Pimephales promelas*). EG & G Bionomics, Wareham, Massachusetts. Laboratory contract number 68-001-0738. Submitted to Environmental Research Laboratory Office of Research and Development, US Environmental Protection Agency, Duluth, Minnesota. EPA contract number EPA-600/3-76-099. USEPA MRID 5008271.

RelevanceReliabilityScore: 92.5Score: 82.5Rating: RRating: R

Relevance points taken off for: Control response (7.5). 100-7.5=92.5

	Macek et al. 1976	D. magna
Parameter	Value	Comment
Test method cited	Standard methods for the	
	estimation of water and	
	wastewater (APHA 1971)	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	<24 h	
phase		
Source of organisms	University of New	
	Hampshire, Durham, New	
	Hampshire	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	No	
Effect 1	Mortality	
Control response 1	Not reported	
Effect 2	Reproduction	
Temperature	20 ± 1 °C	
Test type	Static	

	Macek et al. 1976	D. magna
Parameter	Value	Comment
Photoperiod/light intensity	Continuous/intensity not	
	reported	
Dilution water	Well water	
рН	6.8-7.2	
Hardness	37 mg/L CaCO ₃	
Alkalinity	35 mg/L CaCO ₃	
Dissolved Oxygen	4.6-8.7 mg/L	50-96 %
Feeding	Trout starter	
Purity of test substance	97 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	40-60 %	
Toxicity values calculated based on	Nominal	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acute: Acetone, 43 mg/L	
test solutions		
Concentration 1 Nom; Meas (µg/L)	6; 2.4	4 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	12; 7.2	4 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	25; 14	4 reps, 5/rep
Concentration 4 Nom; Meas (µg/L)	50; 25.6	4 reps, 5/rep
Concentration 5 Nom; Meas (µg/L)	100; 52.7	4 reps, 5/rep
Control	0; 0	4 reps, 5/rep
LC ₅₀ (95% CI) (μg/L)	193 (115-327)	Method: probit

Notes:

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Conductivity (2), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-10=90

<u>Acceptability:</u> Control response (9), Measured concentrations within 20% nominal (4), Carrier solvent (4), Conductivity (1), Random design (2), Adequate replication (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-25 = 75

Reliability score: mean(90, 75)=82.5

Lemna gibba

Study: Fairchild, J.F., Ruessler, D.S., Haverland, P.S. and Carlson, A.R., 1997. Comparative sensitivity of *Selenastrum capricornutum* and *Lemna minor* to sixteen herbicides. Archives of Environmental Contamination and Toxicology, 32(4), 353-357.

RelevanceReliabilityScore: 92.5Score: 75Rating: RRating: R

Relevance points taken off for: Control response (7.5). 100-7.5=92.5

	Fairchild et al. 1997	L. gibba
Parameter	Value	Comment
Test method cited	American Society for	
	Testing and Materials. 1993.	
	Standard guide for	
	conducting static 96h	
	toxicity tests with	
	microalgae: Practice E	
	1218-90. In: Annual book of	
	ASTM standards:Water and	
	environmental	
	technology. ASTM	
	Committee E-47 on	
	Biological Effects	
	and Environmental Fate,	
	American Society for	
	Testing and Materials,	
	Philadelphia, PA, p 929	
Order	Alismatales	
Family	Araceae	
Genus	Lemna	
Species	gibba	
Family native to North America?	Yes	
Age/size at start of test/growth	Not reported	
phase		
Source of organisms	Carolina Biological Supply	Burlington, North
	Company	Carolina
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Yes	
Test duration	96 h	
Data for multiple times?	48, 72, 96 h	

	Fairchild et al. 1997	L. gibba
Parameter	Value	Comment
Effect 1	Biomass	
Control response 1	Not reported	
Temperature	25 °C	
Test type	Static	
Photoperiod/light intensity	16:8 light:dark/400 foot- candle	
Dilution water	Nutrient enriched water, modified from APHA 1985	American Public Health Association, American Water Works Association, and the Water Pollution Control Federation (1985) Standard methods for the examination of water and wastewater, 14th ed., APHA-AWWA- WPCF, Washington, DC.
Feeding	Nutrient enriched water	
Purity of test substance	Technical	
Concentrations measured?	No	
Measured is what % of nominal? Toxicity values calculated based on nominal or measured	Not applicable Nominal	
concentrations? Chemical method documented?	Not applicable	
Concentration of carrier (if any) in	Acetone, concentration not	
test solutions	reported	
Concentration 1 Nom; Meas (µg/L)	Concentrations not reported, 5 concentrations plus solvent and negative controls	3 reps, 12 fronds/rep
Control	Solvent Negative	
EC ₅₀ (95% CI) (μg/L)	170 (10-330)	Method: nonlinear regression
NOEC	75	Method: Duncan's Multiple Range Test p: 0.05 MSD: not reported
LOEC	150	
MATC (GeoMean NOEC, LOEC)	106	

	Fairchild et al. 1997	L. gibba
Parameter	Value	Comment
% control at NOEC	Not calculable	
% control at LOEC	Not calculable	

Notes: Raw data not reported so % controls at NOEC/LOEC not calculable and control responses unknown.

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because the nutrient enriched water used is an industry standard and the medium is presumably appropriate for the test species because a specific water was used.

Reliability points taken off for:

<u>Documentation</u>: Organism life stage/size (5), Nominal concentrations (3), Measured concentrations (3), Statistical significance (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-17 =83

<u>Acceptability:</u> Control response (9),Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Carrier solvent (4), Temperature variation (3), Number of concentrations (3), Dilution factor (2), Hypothesis tests (3). Total: 100- 32=68

Reliability score: mean(83,67)=75

Lemna gibba

Study: Milazzo DP, Servinski MF, Brown RP, Hugo JM, Martin MD. (1993) Trifluralin technical grade 95%: toxicity to the aquatic plant, duckweed, *Lemna gibba* L. G-3. The Environmental Toxicology & Chemistry Research Laboratory, Midland, Michigan. Laboratory project study ID DECO-ES-2653. Submitted to DowElanco, Indianapolis, Indiana. USEPA MRID 42834104.

RelevanceReliabilityScore: 100Score: 96Rating: RRating: R

	Milazzo et al. 1993	L. gibba
Parameter	Value	Comment
Test method cited	Pesticide Assessment Guidelines, Subdivision J Hazard Evaluation: Non- target Plants, USEPA	
Order	Alismatales	
Family	Araceae	
Genus	Lemna	
Species	gibba	
Family native to North America?	Yes	
Age/size at start of test/growth phase	1 w	
Source of organisms	Smithsonian Institution Radiation Biology Laboratory, Rockville, Maryland	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	14 d	
Data for multiple times?	No	
Effect 1	Growth	
Control response 1	Plants: 153 Fronds: 531	
Temperature	25 ± 2 °C	
Test type	Static	
Photoperiod/light intensity	Continuous/5382 lux	
Dilution water	Algal assay medium	
рН	8.3-9.0	
Feeding	Growth medium	

	Milazzo et al. 1993	L. gibba
Parameter	Value	Comment
Purity of test substance	97.9 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	81-102 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	3.13; 2.53	3 reps, 5 plants/rep
Concentration 2 Nom; Meas (µg/L)	6.26; 5.91	
Concentration 3 Nom; Meas (µg/L)	12.6; 12.9	
Concentration 4 Nom; Meas (µg/L)	25.2; 25.3	
Concentration 5 Nom; Meas (µg/L)	50.4; 45.5	
Concentration 6 Nom; Meas (µg/L)	101; 91.3	
Control	Negative: 0; 0	
	Solvent: 0; 0	
EC ₅₀ (95% CI) (μg/L)	Plants: 55.9 (38.96-72.74)	Method:
	Fronds: 53.9 (41.03-66.84)	Numerically
		derived
NOEC	Plants: 12.9	Method: ANOVA
	Fronds: 12.9	and Dunnett's test
		p: 0.05
		MSD: Not reported
LOEC	Plants: 25.3	
MATC	18	
% control at NOEC	Plants: 93 %	Plants: 143 (tmt) /
	Fronds: 96 %	153 (controls) = 93
		Fronds: 508 (tmt) /
		531 (controls) = 96
% control at LOEC	Plants: 75 %	Plants: 115 (tmt) /
		153 (controls) = 75

Notes: Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires Type I water, and the medium is presumably appropriate for the test species because a specific culture media was used.

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2). Total: 100- 2=98

Acceptability: Temperature variation (3), Random design (2), Minimum significant difference

(1). Total: 100-6 = 94

Reliability score: mean(98,94)=96

Navicula pelliculosa

Study: Hughes JS, Williams TL. (1993b) The toxicity of trifluralin to *Navicula pelliculosa*. Malcolm Pirnie, Inc., Tarrytown, New York. Laboratory study number B460-153-3. Submitted to The Dow Chemical Company, Indianapolis, Indiana. USEPA MRID 42834102.

RelevanceReliabilityScore: 100Score: 96Rating: RRating: R

	Hughes & Williams 1993b	N. pelliculosa
Parameter	Value	Comment
Test method cited	Pesticide Assessment	
	Guidelines, USEPA	
Phylum/subphylum	Heterokontophyta	
Class	Bacillariophyceae	
Order	Naviculales	
Family	Naviculaceae	
Genus	Navicula	
Species	pelliculosa	
Family native to North America?	Yes	
Age/size at start of test/growth	Algal cells	
phase		
Source of organisms	Laboratory cultures	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	Given organism
		size and presence in
		growth medium, it
		is assumed that
		aliquots are
		inherently randomly
Test vessels randomized?	Not reported	
Test duration	5 d	
Data for multiple times?	3, 4, 5 d	
Effect 1	Cell count	
Control response 1	806,590	
Temperature	24 ± 2 °C	
Test type	Static	
Photoperiod/light intensity	Continuous/400 footcandles	
Dilution water	Synthetic AAP/Si medium	Made with Type I water
pH	7.5	114101
Feeding	Nutrient medium	
1 ccumg	1 Tuti Ciit iii Cuiuiii	

	Hughes & Williams 1993b	N. pelliculosa
Parameter	Value	Comment
Purity of test substance	97.92 %	
Concentrations measured?	Measured	
Measured is what % of nominal?	100-121 %	
Toxicity values calculated based on nominal or measured	Measured	Initial values
concentrations?		
Chemical method documented?	HPLC	
Concentration of carrier (if any) in test solutions	Dimethylformamide	
Concentration 1 Nom; Meas (µg/L)	6.36; 7.65	4 reps, 3,000 cells/rep
Concentration 2 Nom; Meas (µg/L)	12.7; 15.4	4 reps, 3,000 cells/rep
Concentration 3 Nom; Meas (µg/L)	25.3; 25.3	4 reps, 3,000 cells/rep
Concentration 4 Nom; Meas (µg/L)	50.6; 54.3	4 reps, 3,000 cells/rep
Concentration 5 Nom; Meas (µg/L)	101; 118	4 reps, 3,000 cells/rep
Concentration 6 Nom; Meas (µg/L)	202; 238	4 reps, 3,000 cells/rep
Control	Negative: 0; 0 Solvent: 0; 0	4 reps, 3,000 cells/rep
EC ₅₀ (95% CI) (μg/L)	15.3 (6.72-34.7)	Method: Non-linear regression
NOEC	<7.65	Method: ANOVA, Dunnett's test p: 0.05 MSD: Not reported
% control at NOEC	Not calculable	1

Notes: Based on initial (0 d) measured values; by day 5 no detectable amounts of trifluralin in any treatment. The chemical is not stable under these conditions. This is likely due to photolytic degradation.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires Type I water, and the medium is presumably appropriate for the test species because a specific culture media was used. Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2). Total: 100-2 = 98

Acceptability: Temperature variation (3), Random design (2), Minimum significant difference

(1). Total: 100-6 = 94

Reliability score: mean(98, 94)=96

Oncorhynchus mykiss

Study: Adams ER, Cocke PJ, Gunnoe MD. (1990) The toxicity of trifluralin to rainbow trout (*Salmo gairdneri*) in a 48-day life-stage study. Lilly Research Laboratories, Greenfield, Indiana. Laboratory project identification F02489. USEPA MRID 41386202.

RelevanceReliabilityScore: 100Score: 92Rating: RRating: R

	Adams et al. 1990	O. mykiss
Parameter	Value	Comment
Test method cited	Pesticide Assessment	
	Guidelines, Subdivision E,	
	Hazard Evaluation: Wildlife	
	and Aquatic Organisms,	
	USEPA 1982; Hazard	
	Evaluation Division	
	Standard Evaluation	
	Procedure, Fish Early Life-	
	Stage Test, USEPA 1986;	
	Standard Guide for	
	Conducting Early Life-	
	Stage Toxicity Tests with	
	Fishes, ASTM, 1988	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Salmoniformes	
Family	Salmonidae	
Genus	Oncorhynchus	
Species	mykiss	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Eye stage eggs	
Source of organisms	Trout Lodge, McMillin,	
C	Washington	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Yes	
Test duration	48 d	
Data for multiple times?	No	
Effect 1	Time to hatch	
Control response 1	9.1 d	
Effect 2	Survival	

	Adams et al. 1990	O. mykiss
Parameter	Value	Comment
Control response 2	83.3 %	
Effect 3	Growth	
Control response 3	Length: 36.5 mm	
_	Weight: 0.47 g	
Temperature	12.7 ± 0.2 °C	
Test type	Flow-through	
Photoperiod/light intensity	16:8, 1:d/ \leq 22 μ E/m ² /s	
Dilution water	Well water	
pН	7.4-8.5	
Hardness	128 mg/L CaCO ₃	
Alkalinity	153 mg/L CaCO ₃	
Conductivity	196 μS/cm	
Dissolved Oxygen	10 mg/L	94 %
Feeding	Salmon starter mesh, 2-3/d	
Purity of test substance	99.86 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	57-62 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acetone, 0.02 mL/L	
test solutions		
Concentration 1 Nom; Meas (µg/L)	0.95; 0.59	4 reps, 20/rep
Concentration 2 Nom; Meas (µg/L)	1.9; 1.14	4 reps, 20/rep
Concentration 3 Nom; Meas (µg/L)	3.8; 2.18	4 reps, 20/rep
Concentration 4 Nom; Meas (µg/L)	7.6; 4.32	4 reps, 20/rep
Concentration 5 Nom; Meas (µg/L)	15.2; 8.81	4 reps, 20/rep
Control	Negative: 0	4 reps, 20/rep
	Solvent: 0	
NOEC	1.14	Method: Weighted
		ANOVA
		p: 0.05
		MSD: Not reported
		Based on survival
LOEC	8.81	Based on survival
% control at NOEC	Time to hatch = 93 %	Time to hatch: 8.5
	Survival = 106 %	(tmt) / 9.1 (mean
	Length: 98 %	controls) = 93
	Weight: 94 %	Survival: 88.6 (tmt)
		/ 83.3 (mean
		controls) = 106
		Length: 35.9 (tmt) /
		36.5 (mean
		controls) = 98
		Weight: 0.44 (tmt) /

	Adams et al. 1990	O. mykiss
Parameter	Value	Comment
		0.47 (mean
		controls) = 94

Notes:

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2), Point estimates (8). Total: 100-10 =90

<u>Acceptability:</u> Organisms randomized (1), Minimum significant difference (1), % control at LOEC (1), Point estimates (3). Total: 100-6 =94

Reliability score: mean(90,94)=92

Pimephales promelas

Study: Macek KJ, Lindberg MA, Sauter S, Buxton KS and Costa PA. (1976) Toxicity of Four Pesticides to Water Fleas and Fathead Minnows: Acute and Chronic Toxicity of Acrolein, Heptachlor, Endosulfan, and Trifluralin to the Water Flea (*Daphnia magna*) and the Fathead Minnow (*Pimephales promelas*). EG & G Bionomics, Wareham, Massachusetts. Laboratory contract number 68-001-0738. Submitted to Environmental Research Laboratory Office of Research and Development, US Environmental Protection Agency, Duluth, Minnesota. EPA contract number EPA-600/3-76-099. USEPA MRID 5008271.

RelevanceReliabilityScore: 100Score: 91Rating: RRating: R

Relevance points taken off for: none

	Macek et al. 1976	P. promelas
Parameter	Value	Comment
Test method cited	Standard methods for the	
	estimation of water and	
	wastewater (APHA 1971)	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Pimephales	
Species	promelas	
Family native to North America?	Yes	
Age/size at start of test/growth	26 d	
phase		
Source of organisms	Newtown Fish Toxicology	
	Laboratory, Newtown, Ohio	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	Acute: 48 h	
	Chronic: 61 w	
Data for multiple times?	30, 60 d	
	61 w	
Effect 1	Survival	
Control response 1	Generation 1:	
	30 d: 100 %	
	60 d: 100 %	
	61 w: 83.5 %	

	Macek et al. 1976	P. promelas
Parameter	Value	Comment
	Generation 2 (fry):	
	30 d: 84 %	
	60 d: 74 %	
Effect 2	Length	
Control response 2	Generation 1:	
1	30 d: 21 mm	
	60 d: 24.5 mm	
	Generation 2:	
	30 d: 17 mm	
	60 d: 22 mm	
Temperature	25 ± 1 °C	
Test type	Static renewal	Intermittent flow
Photoperiod/light intensity	Standard day	
	length/intensity not reported	
Dilution water	Well water	
рН	6.6-7.2	
Hardness	33 mg/L CaCO ₃	
Alkalinity	27 mg/L CaCO ₃	
Dissolved Oxygen	5.9-9.5 mg/L	65-105 %
Feeding	Ad libitum 2/d trout starter,	
	daphnids, and brine shrimp	
	naulpii	
Purity of test substance	97 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	76-125 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acute: Acetone, 12 mg/L	
test solutions	Chronic: none	
Concentration 1 Nom; Meas (µg/L)	1.2; 1.5	2 reps, 40/rep
Concentration 2 Nom; Meas (µg/L)	2.5; 1.9	•
Concentration 3 Nom; Meas (µg/L)	5; 5.1	
Concentration 4 Nom; Meas (µg/L)	10; 8.2	
Concentration 5 Nom; Meas (µg/L)	20; 16.5	
Control	Negative, 0; 0	
LC ₅₀ (95% CI) (μg/L)	115	Method:
NOEC (95% CI) (μg/L)		Method:
NOEC	1.9 µg/L	
		p: MSD:
LOEC	5.1 μg/L	141010.
MATC (GeoMean NOEC, LOEC)	3.1 µg/L	
WITTE (Georgean NOEC, LOEC)	3.1	
% control at NOEC	Survival:	Survival:
	60 d: 97.5 %	60 d: 97.5 (tmt) /
	00 d . 71.3 /0	00 d. 77.3 (tillt)/

	Macek et al. 1976	P. promelas
Parameter	Value	Comment
	Length:	100 (control) = 97.5
	60 d: 106 %	Length:
		60 d: 26 (tmt) / 24.5
		(control) = 106
% control at LOEC	Survival:	Survival:
	60 d: 96.5 %	60 d: 96.5 (tmt) /
	Length:	100 (control) = 96.5
	60 d: 100 %	Length:
		60 d: 24.5 (tmt) /
		24.5 (control) =
		100

Notes: All chronic exposure tanks flushed with 3.7 g/L malachite green and 25 μ g/L formalin between days 115-130 to remove external parasites.

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Conductivity (2), Minimum significant difference (2). Total: 100-4 = 96

<u>Acceptability:</u> Measured concentrations within 20% nominal (4), Carrier solvent (4), Conductivity (1), Random design (2), Adequate replication (2), Minimum significant difference (1). Total: 100-14 = 86

Reliability score: mean(96,86)=91

Pimephales promelas

Study: Macek KJ, Lindberg MA, Sauter S, Buxton KS and Costa PA. (1976) Toxicity of Four Pesticides to Water Fleas and Fathead Minnows: Acute and Chronic Toxicity of Acrolein, Heptachlor, Endosulfan, and Trifluralin to the Water Flea (*Daphnia magna*) and the Fathead Minnow (*Pimephales promelas*). EG & G Bionomics, Wareham, Massachusetts. Laboratory contract number 68-001-0738. Submitted to Environmental Research Laboratory Office of Research and Development, US Environmental Protection Agency, Duluth, Minnesota. EPA contract number EPA-600/3-76-099. USEPA MRID 5008271.

RelevanceReliabilityScore: 92.5Score: 82.5Rating: RRating: R

Relevance points taken off for: Control response (7.5). 100-7.5=92.5

	Macek et al. 1976	P. promelas
Parameter	Value	Comment
Test method cited	Standard methods for the	
	estimation of water and	
	wastewater (APHA 1971)	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Pimephales	
Species	promelas	
Family native to North America?	Yes	
Age/size at start of test/growth	26 d	
phase		
Source of organisms	Newtown Fish Toxicology	
_	Laboratory, Newtown, Ohio	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	Acute: 48 h	
Data for multiple times?	Not reported	
Effect 1	Mortality	
Control response 1	Not reported	
Temperature	25 ± 1 °C	
Test type	Static renewal	Intermittent flow
Photoperiod/light intensity	Standard day	
	length/intensity not reported	

	Macek et al. 1976	P. promelas
Parameter	Value	Comment
Dilution water	Well water	
pH	6.6-7.2	
Hardness	33 mg/L CaCO ₃	
Alkalinity	27 mg/L CaCO ₃	
Dissolved Oxygen	5.9-9.5 mg/L	65-105 %
Feeding	Ad libitum 2/d trout starter,	
	daphnids, and brine shrimp	
	naulpii	
Purity of test substance	97 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	76-125 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acute: Acetone, 12 mg/L	
test solutions		
Concentration 1 Nom; Meas (µg/L)	1.2; 1.5	2 reps, 40/rep
Concentration 2 Nom; Meas (µg/L)	2.5; 1.9	
Concentration 3 Nom; Meas (µg/L)	5; 5.1	
Concentration 4 Nom; Meas (µg/L)	10; 8.2	
Concentration 5 Nom; Meas (µg/L)	20; 16.5	
Control	Negative, 0; 0	
LC ₅₀ (95% CI) (μg/L)	115 (48-211)	Method: probit

Notes:

Trifluralin solubility (S) = 238.08 $\mu g/L$, 2S = 476.16 $\mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Conductivity (2), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100- 10=90

<u>Acceptability:</u> Test response (9), Measured concentrations within 20% nominal (4), Carrier solvent (4), Conductivity (1), Random design (2), Adequate replication (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100- 25=75

Reliability score: mean(90,75)=82.5

Raphidocelis subcapitata

Study: Adams ER and Cocke PJ. (1990) Toxicity of trifluralin to a freshwater alga (*Selenastrum capricornutum*) in a static test system. Lilly Research Laboratories, Greenfield, Indiana. Laboratory project identification J00989. USEPA MRID 41934502.

RelevanceReliabilityScore: 100Score: 95.5Rating: RRating: R

Relevance points taken off for: None.

	Adams &Cocke 1990	R. subcapitata
Parameter	Value	Comment
Test method cited Phylum/subphylum	Algal, Growth Inhibition Test, OECD 1984; Pesticide Assessment Guidelines, Subdivision J, Hazard Evaluation: Nontarget Plants, USEPA 1982. Chlorophyta	
Class	Chlorophyceae	
Order	Sphaeropleales	
Family	Selenastraceae	
Genus	Raphidocelis	
Species	subcapitata	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Algal cells	
Source of organisms	Laboratory cultures	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	Given organism size and presence in growth medium, it is assumed that aliquots are inherently randomly
Test vessels randomized?	Not reported	
Test duration	7 d	
Data for multiple times?	1, 2, 3, 4, 5, 7 d	
Effect 1	Growth rate	
Control response 1 (mean controls)	0.551/d	
Effect 2	Cell count	
Control response 2 (mean controls)	$3295 \times 10^3 / \text{mL}$	

	Adams &Cocke 1990	R. subcapitata
Parameter	Value	Comment
Effect 3	Biomass	
Control response 3 (mean controls)	0.070 mg/mL	
Temperature	$22.5 \pm 0.5 ^{\circ}\text{C}$	
Test type	Static	
Photoperiod/light intensity	Continuous/4 klux	
Dilution water	Nutrient medium	Made with deionized ultra- filtered sterilized water
рН	7.6-7.8	
Hardness	51 mg/L CaCO ₃	
Alkalinity	20 mg/L CaCO ₃	
Conductivity	109 μS/cm	
Dissolved Oxygen	Not reported	
Feeding	Nutrient medium	
Purity of test substance	99.86 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	0d: 87-102 % 7d: 7-21 %	Rapid dissipation due to volatilization and photolytic degradation
Toxicity values calculated based on nominal or measured concentrations? Chemical method documented?	Measured	
Concentration of carrier (if any) in test solutions	Acetone, 10 μL/L	
Concentration 1 Nom; Meas (µg/L)	10; 2.12	3 reps, 10,000 cells/rep
Concentration 2 Nom; Meas (µg/L)	80; 5.37	3 reps, 10,000 cells/rep
Concentration 3 Nom; Meas (µg/L)	160; 11.7	3 reps, 10,000 cells/rep
Concentration 4 Nom; Meas (µg/L)	320; 21.9	3 reps, 10,000 cells/rep
Concentration 5 Nom; Meas (µg/L)	640; 62.1	3 reps, 10,000 cells/rep
Control	Negative: 0 Solvent: 0	1
EC ₅₀ (95% CI) (μg/L)	12.2	Method: Linear regression
NOEC	5.37	Method: Dunnett's t-test p: MSD:
LOEC	11.7	

	Adams & Cocke 1990	R. subcapitata
Parameter	Value	Comment
MATC	7.9	
% control at NOEC	Growth rate: 95 % Cell count: 60 % Biomass: 77 %	Growth rate: 0.526 (tmt) / 0.551 (mean controls) = 95 Cell count: 1970 (tmt) / 3295 (mean controls) = 60 Biomass: 0.054 (tmt) / 0.070 (mean controls) = 77
% control at LOEC	Growth rate: 71 % Cell count: 60 % Biomass: 29 %	Growth rate: 0.392 (tmt) / 0.551 (mean controls) = 71 Cell count: 1970 (tmt) / 3295 (mean controls) = 60 Biomass: 0.02(tmt) / 0.070 (mean controls) = 29

Notes: Dissolved oxygen not reported but water quality points not deducted because oxygen-producing alga.

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2). Total: 100-2 = 98

<u>Acceptability:</u> Measured concentrations within 20% nominal (4), Random design (2), Minimum significant difference (1). Total: 100-7 =93

Reliability score: mean(98, 93)=95.5

Raphidocelis subcapitata

Study: Fairchild, J.F., Ruessler, D.S., Haverland, P.S. and Carlson, A.R., 1997. Comparative sensitivity of *Selenastrum capricornutum* and *Lemna minor* to sixteen herbicides. Archives of Environmental Contamination and Toxicology, 32(4), 353-357.

RelevanceReliabilityScore: 92.5Score: 75.5Rating: RRating: R

Relevance points taken off for: Control response (7.5). 100-7.5=92.5

	Fairchild et al. 1997	R. subcapitata
Parameter	Value	Comment
Test method cited	American Society for	
	Testing and Materials. 1993.	
	Standard guide for	
	conducting static 96h	
	toxicity tests with	
	microalgae: Practice E	
	1218-90. In: Annual book of	
	ASTM standards:Water and	
	environmental	
	technology. ASTM	
	Committee E-47 on	
	Biological Effects	
	and Environmental Fate,	
	American Society for	
	Testing and Materials,	
	Philadelphia, PA, p 929	
Phylum/subphylum	Chlorophyta	
Class	Chlorophyceae	
Order	Sphaeropleales	
Family	Selenastraceae	
Genus	Raphidocelis	
Species	subcapitata	
Family native to North America?	Yes	
Age/size at start of test/growth	Not reported	
phase		
Source of organisms	Carolina Biological Supply	Burlington, North
	Company	Carolina
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	Given organism
		size and presence in

	Fairchild et al. 1997	R. subcapitata
Parameter	Value	Comment
		growth medium, it
		is assumed that
		aliquots are
		inherently randomly
Test vessels randomized?	Yes	
Test duration	96 h	
Data for multiple times?	48, 72, 96 h	
Effect 1	Biomass	
Control response 1	Not reported	
Temperature	25 °C	
Test type	Static	
Photoperiod/light intensity	16:8 light:dark/400 foot-	
	candle	
Dilution water	ASTM growth medium	
Feeding	Growth medium	
Purity of test substance	Technical	
Concentrations measured?	No	
Measured is what % of nominal?	Not applicable	
Toxicity values calculated based on	Nominal	
nominal or measured		
concentrations?		
Chemical method documented?	Not applicable	
Concentration of carrier (if any) in	Acetone, concentration not	
test solutions	reported	
Concentration 1 Nom; Meas (µg/L)	Concentrations not reported,	3 reps, 20,000
	5 concentrations plus	cells/mL/rep
	solvent and negative	
	controls	
Control	Solvent	
	Negative	
EC ₅₀ (95% CI) (μg/L)	673 (594-751) > 2S	Method: nonlinear
11070	1.70	regression
NOEC	150	Method: Duncan's
		Multiple Range
		Test
		p: 0.05
LOEG	200	MSD: not reported
LOEC NATE (G. M. NOEG LOEG)	300	
MATC (GeoMean NOEC, LOEC)	212.1	
% control at NOEC	Not calculable	
% control at LOEC	Not calculable	
Notes: Pary data not reported so 0/ ac	· 1 · NOEGLOEG · 1	1 1 1 1 1 1

Notes: Raw data not reported so % controls at NOEC/LOEC not calculable and control responses unknown.

Trifluralin solubility (S) = 238.08 $\mu g/L$, 2S = 476.16 $\mu g/L$.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used is an ASTM standard for this species, and the medium is presumably appropriate for the test species because a specific culture media was used.

Reliability points taken off for:

<u>Documentation</u>: Organism life stage/size (5), Nominal concentrations (3), Measured concentrations (3), Statistical significance (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-17 =83

<u>Acceptability:</u> Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Carrier solvent (4), Temperature variation (3), Number of concentrations (3), Dilution factor (2), Hypothesis tests (3). Total: 100- 32=68

Reliability score: mean(83,68)=75.5

Appendix A2 – Wildlife Toxicity Studies Rated R

A. platyrhynchos

Study: Beavers JB, Dukes V, Jaber MJ. (1987) Trifluralin technical: a one-generation reproduction study with the mallard (*Anus platyrhynchos*). Wildlife International Limited, Easton, Maryland. Laboratory project number 228-102. Submitted to Industria Prodotti Chimici, Novate Milanese, Italy. USEPA MRID 40334704.

Table X.x Documentation and acceptability rating for terrestrial laboratory/field data (adapted from ECOTOX 2006). Score is given if

parameter is reported.

Parameter ¹	Score ²	Points
Exposure duration	20	20
Control type	7	7
Organism information (i.e., age, life stage)	8	8
Chemical grade or purity	5	5
Chemical analysis method	5	5
Exposure type (i.e., dermal, dietary, gavage)	10	10
Test location (i.e., laboratory, field, natural artificial)	5	5
Application frequency	5	5
Organism source	5	5
Organism number and/or sample number	5	5
Dose number	5	5
Statistics		
Hypothesis tests		
Statistical significance	5	5
Significance level	5	0
Minimum significant difference	3	0
% of control at NOEC and/or LOEC	3	0
Point estimates (i.e., LC ₅₀ , EC ₅₀)	4	0
Total	100	85

¹ Compiled from RIVM (2001), USEPA (1985; 2003b), ECOTOX (2006), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

² Weighting based acceptability criteria from various ASTM, OECD, APHA, and USEPA methods, ECOTOX (2006), and on data quality criteria in RIVM (2001), USEPA (1985; 2003b), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

Appendix A3 – Studies rated RL, LR, LL

Anaxyrus fowleri

Study: Sanders, H.O., 1970. Pesticide toxicities to tadpoles of the western chorus frog Pseudacris triseriata and Fowler's toad Bufo woodhousii fowleri. *Copeia*, 246-251.

RelevanceReliabilityScore: 75Score: 61Rating: LRating: L

Relevance points taken off for: Standard method (10), Controls (15). 100-25=75

	Sanders 1970	A. fowleri
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata/Vertebrata	
Class	Amphibia	
Order	Anura	
Family	Bufonidae	
Genus	Anaxyrus	
Species	fowleri	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Tadpoles	
Source of organisms	Ponds near Fish-Pesticide	
200200 or organisms	Research Laboratory,	
	Columbia Missouri	
Have organisms been exposed to	Not reported	
contaminants?		
Animals acclimated and disease-	Eggs hatched in lab,	
free?	tadpoles acclimated 2 h	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 96 h	
Effect 1	Mortality	
Control response 1	Not reported	
Temperature	$15.5 \pm 0.5 ^{\circ}\text{C}$	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted demineralized	
	water	
рН	7.1	
Hardness	Not reported	
Alkalinity	30 mg/L	
Conductivity	Not reported	
Dissolved Oxygen	mg/L	

	Sanders 1970	A. fowleri
Parameter	Value	Comment
Feeding		
Purity of test substance	Technical	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	Ethanol, concentration not	
test solutions	reported	
Concentration 1 Nom; Meas (µg/L)	Concentrations nor	reps, 10/rep
	reported; 4-5 used with	
	controls	
LC ₅₀ (95% CI) (μg/L)	24 h: 180 (100-300)	Method: Modified
	48 h: 170 (100-310)	Litchfield and
	96 h: 100 (80-490)	Wilcoxon

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Nominal concentrations (3), Measured concentrations (3), Hardness (2), Conductivity (2), Temperature (4), Photoperiod (3), Hypothesis tests (8). Total: 100-33=67

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations (4), Concentrations not > 2x solubility (4), Carrier solvent (4), No prior contamination (4), Organisms randomized (1), Hardness (2), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Hypothesis tests (3). Total: 100-45 =55

Reliability score: mean(67,55)=61

Carrassius auratus

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

RelevanceReliabilityScore: 75Score: 60Rating: LRating: L

Relevance points taken off for: Standard method (10), Controls (15). 100-25=75

	Johnson & Finley 1980	C. auratus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Carrassius	
Species	auratus	
Family native to North America?	Yes	
Age/size at start of test/growth	1.0 g	
phase		
Source of organisms	Hatchery	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	18 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
pН	7.2-7.5	
Hardness	272 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	

	Johnson & Finley 1980	C. auratus
Parameter	Value	Comment
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	145 (108-195)	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Cyprinodon variegatus

Study: Parrish PR, Dyer EE, Enos JM, Wilson WG. (1978) Chronic toxicity of chlordane, trifluralin, and pentachlorophenol to Sheepshead Minnows (*Cyprinodon variegatus*). EG&G Bionomics Marine Research Laboratory, Pensacola, Florida. Laboratory project study ID EPA-600/3-78-010. Submitted to Environmental Research Laboratory, USEPA, Gulf Breeze, Florida. USEPA MRID 42449901.

RelevanceReliabilityScore: 85Score: 84.5Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100-15=85

	Parrish et al. 1978	C. variegatus
Parameter	Value	Comment
Test method cited	The Committee on Methods	
	for Toxicity Tests with	
	Aquatic Organisms	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cyprinodontiformes	
Family	Cyprinodontidae	
Genus	Cyprinodon	
Species	variegatus	
Family native to North America?	Yes	
Age/size at start of test/growth	Fry	
phase		
Source of organisms	Collected near laboratory	
Have organisms been exposed to	Not reported	
contaminants?		
Animals acclimated and disease-	Yes, 14 d before collecting	
free?	eggs/testes for fertilization	
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	166 d	
Data for multiple times?	Various	
Effect 1	Mortality	
Control response 1	Negative control:	
	1-40 d: 9	
	41-80 d: 0	
	81-120 d: 2	
	121-166 d: 0	
	Solvent control:	
	1-40 d: 5	
	41-80 d: 0	
	81-120 d: 0	

	Parrish et al. 1978	C. variegatus
Parameter	Value	Comment
	121-166 d: 0	
Effect 2	Growth	
Control response 2, mean controls	Length, 166 d: 3.4	
-	Weight, 166 d: 1.05	
Effect 3	Eggs spawned	
Control response 3	Negative control:	
	113-122 d: 839	
	136-145 d: 684	
	157-166 d: 489	
	Solvent control:	
	113-122 d: 839	
	136-145 d: 318	
	157-166 d: 684	
Temperature	30 ± 1 °C	
Test type	Intermittent flow	
Photoperiod/light intensity	16:8, 1:d/1,100 lux	
Dilution water	Natural seawater	Filtered and aerated
Dissolved Oxygen	Not reported	
Feeding	Live brine shrimp nauplii or	
-	flaked commercial fish food	
	ad libitum	
Purity of test substance	99 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	21-38%	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	6.3; 1.3	10 reps, 20/rep
Concentration 2 Nom; Meas (µg/L)	12.5; 4.8	10 reps, 20/rep
Concentration 3 Nom; Meas (µg/L)	25.; 9.6	10 reps, 20/rep
Concentration 4 Nom; Meas (µg/L)	50; 17.7	10 reps, 20/rep
Concentration 5 Nom; Meas (µg/L)	100; 34.1	10 reps, 20/rep
Control	Negative: 0; 0	10 reps, 20/rep
	Solvent: 0; 0	
LC ₅₀ (95% CI) (µg/L)	10 d: 84 (48-145)	Method: Probit,
- 30 (x - / / (p 0 / - /	Incipient value	linear regression
NOEC	1.3	Method: ANOVA
		p: 0.05
		MSD:
LOEC	4.8	
MATC	2.5	
% control at NOEC	Mortality: 35 %	Mortality,
	Length: 103 %	cumulative:

	Parrish et al. 1978	C. variegatus
Parameter	Value	Comment
	Weight: 109 %	166 d: 6 (tmt) / 17
	Eggs spawned:	(mean controls) =
	113-122 d: 65 %	35
	136-145 d: 60 %	Length, 166 d: 3.5
	157-166 d: 83 %	(tmt) / 3.4 (mean
		controls) = 103
		Weight, 166 d: 1.2
		(tmt) / 1.1 (mean
		controls) = 109
		Eggs spawned: 113-
		122 d: 377 (tmt) /
		578 (mean controls)
		= 65
		136-145 d: 418
		(tmt) / 696 (mean
		controls) = 60
		157-166 d: 515
		(tmt) / 619 (mean
		controls) = 83
% control at LOEC	Mortality: 165 %	Mortality,
	Length: 94 %	cumulative:
	Weight: 91 %	28 (tmt) / 17 (mean
	Eggs spawned:	controls) = 165
	113-122 d: 75 %	Length, 166 d: 3.2
	136-145 d: 32 %	(tmt) / 3.4 (mean
	157-166 d: 68 %	controls) = 94
		Weight, 166 d: 1
		(tmt) / 1.1 (mean
		controls) = 91
		Eggs spawned:
		113-122 d: 431
		(tmt) / 578 (mean
		controls) = 75
		136-145 d: 222
		(tmt) / 696 (mean
		controls) = 32
		157-166 d: 424
		(tmt) / 619 (mean
		controls) = 68

Notes: Application factor (AF) 0.04-0.06.

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:
Documentation: Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), pH (3), Minimum significant difference (2). Total: 100-15 =85

<u>Acceptability:</u> Hardness (2), Alkalinity (2), Dissolved oxygen (6), Conductivity (1), pH (2), Random design (2), Minimum significant difference (1). Total: 100-=

Reliability score: mean(85,84)=84.5

Cyprinodon variegatus

Study: Parrish PR, Dyer EE, Enos JM, Wilson WG. (1978) Chronic toxicity of chlordane, trifluralin, and pentachlorophenol to Sheepshead Minnows (*Cyprinodon variegatus*). EG&G Bionomics Marine Research Laboratory, Pensacola, Florida. Laboratory project study ID EPA-600/3-78-010. Submitted to Environmental Research Laboratory, USEPA, Gulf Breeze, Florida. USEPA MRID 42449901.

RelevanceReliabilityScore: 85Score: 83.5Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100-15=85

	Parrish et al. 1978	C. variegatus
Parameter	Value	Comment
Test method cited	The Committee on Methods	
	for Toxicity Tests with	
	Aquatic Organisms	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cyprinodontiformes	
Family	Cyprinodontidae	
Genus	Cyprinodon	
Species	variegatus	
Family native to North America?	Yes	
Age/size at start of test/growth	Fry	
phase	1-1.5 cm	
Source of organisms	Collected near laboratory	
Have organisms been exposed to	Not reported	
contaminants?		
Animals acclimated and disease-	Yes, 14 d before collecting	
free?	eggs/testes for fertilization	
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	No	
Effect 1	Mortality	
Control response 1	0 %	
Temperature	30 ± 1 °C	
Test type	Intermittent flow	
Photoperiod/light intensity	16:8, l:d/1,100 lux	
Dilution water	Natural seawater	Filtered and aerated
Dissolved Oxygen	Not reported	

	Parrish et al. 1978	C. variegatus
Parameter	Value	Comment
Feeding	Live brine shrimp nauplii or	
	flaked commercial fish food	
	ad libitum	
Purity of test substance	99 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	47-56 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in		
test solutions		
Concentration 1 Nom; Meas (µg/L)	107; 54	Reps not reported,
		20/rep
Concentration 2 Nom; Meas (µg/L)	142; 76	Reps not reported,
		20/rep
Concentration 3 Nom; Meas (µg/L)	190; 105	Reps not reported,
		20/rep
Concentration 4 Nom; Meas (µg/L)	253; 131	Reps not reported,
		20/rep
Concentration 5 Nom; Meas (µg/L)	337; 189	Reps not reported,
		20/rep
Concentration 6 Nom; Meas (µg/L)	450; 213	Reps not reported,
		20/rep
Concentration 7 Nom; Meas (µg/L)	600; 318	Reps not reported,
		20/rep
Control	Negative: 0; 0	Reps not reported,
		20/rep
LC ₅₀ (95% CI) (μg/L)	190 (128-282)	Method: Probit,
		linear regression

Notes: Application factor (AF) 0.04-0.06.

Trifluralin solubility (S) = 238.08 $\mu g/L$, 2S = 476.16 $\mu g/L$. All measured test exposure concentrations were acceptable.

Reliability points taken off for:

<u>Documentation:</u> Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), pH (3), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-15 =85

<u>Acceptability:</u> Hardness (2), Alkalinity (2), Dissolved oxygen (6), Conductivity (1), pH (2), Random design (2), Adequate replication (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-20 =80

Reliability score: mean(85,82)=83.5

Lepomis macrochirus

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

RelevanceReliabilityScore: 75Score: 60Rating: LRating: L

Relevance points taken off for: Standard method (10), Controls (15). 100-25=75

	Johnson & Finely 1980	L. macrochirus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Perciformes	
Family	Centrarchidae	
Genus	Lepomis	
Species	macrochirus	
Family native to North America?	Yes	
Age/size at start of test/growth	0.8 g	
phase		
Source of organisms	Hatchery	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	22 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
pH	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	

	Johnson & Finely 1980	L. macrochirus
Parameter	Value	Comment
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	58 (47-70)	Method: Litchfield
. <u>-</u>		and Wilcoxon

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Micropterus salmoides

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

RelevanceReliabilityScore: 75Score: 60Rating: LRating: L

Relevance points taken off for: Standard method (10), Controls (15). 100-25=75

	Johnson & Finley 1980	M. salmoides
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Perciformes	
Family	Centrachidae	
Genus	Micropterus	
Species	salmoides	
Family native to North America?	Yes	
Age/size at start of test/growth	0.7 g	
phase		
Source of organisms	Hatchery	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	18 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
рН	7.2-7.5	
Hardness	272 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	

	Johnson & Finley 1980	M. salmoides
Parameter	Value	Comment
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	75 (65-87)	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Oncorhynchus mykiss

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

RelevanceReliabilityScore: 75Score: 60Rating: LRating: L

Relevance points taken off for: Standard method (10), Controls (15). 100-25=75

	Johnson & Finley 1980	O. mykiss
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Salmoniformes	
Family	Salmonidae	
Genus	Oncorhynchus	
Species	mykiss	
Family native to North America?	Yes	
Age/size at start of test/growth	0.8 g	
phase		
Source of organisms	Hatchery	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	12 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
рН	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	

	Johnson & Finley 1980	O. mykiss
Parameter	Value	Comment
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	41 (26-62)	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Pimephales promelas

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

RelevanceReliabilityScore: 75Score: 60Rating: LRating: L

Relevance points taken off for: Standard method (10), Controls (15). 100-25=75

	Johnson & Finley 1980	P. promelas
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Pimephales	
Species	promelas	
Family native to North America?	Yes	
Age/size at start of test/growth	0.8 g	
phase		
Source of organisms	Hatchery	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	18 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
pH	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	

	Johnson & Finley 1980	P. promelas
Parameter	Value	Comment
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	105 (83-134)	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Skeletonema costatum

Study: Hughes JS, Williams TL. (1993c) The toxicity of trifluralin to *Skeletonema costatum*. Malcolm Pirnie, Inc., Tarrytown, New York. Laboratory study number B460-153-3. Submitted to The Dow Chemical Company, Indianapolis, Indiana. USEPA MRID 42834101.

RelevanceReliabilityScore: 85Score: 96Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100-15=85

	Hughes & Williams 1993c	S. costatum
Parameter	Value	Comment
Test method cited	Pesticide Assessment	
	Guidelines, USEPA	
Phylum/subphylum	Bacillariophyta	
Class	Coscinodiscophyceae/	
	Thalassiosirophycidae	
Order	Thalassiosirales	
Family	Skeletonemaceae	
Genus	Skeletonema	
Species	costatum	
Family native to North America?	Yes	
Age/size at start of test/growth	Algal cells	
phase		
Source of organisms	Laboratory stock cultures	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	Given organism
		size and presence in
		growth medium, it
		is assumed that
		aliquots are
		inherently randomly
Test vessels randomized?	Not reported	
Test duration	5 d	
Data for multiple times?	3, 4, 5 d	
Effect 1	Cell count	
Control response 1	5 d: 408,000	
Temperature	20 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	14:10, 1:d/400 footcandles	
Dilution water	Marine algal assay nutrient	Made with ASTM

	Hughes & Williams 1993c	S. costatum
Parameter	Value	Comment
	medium	Type I water
pH	8.24	Mean
Feeding	Nutrient medium	
Purity of test substance	97.92	
Concentrations measured?	Measured	
Measured is what % of nominal?	73-146 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	HPLC	
Concentration of carrier (if any) in	N,N-dimethylformamide,	
test solutions	0.5 mL/L	
Concentration 1 Nom; Meas (µg/L)	3.13; 2.54	3 reps, 10,000 cells/rep
Concentration 2 Nom; Meas (µg/L)	6.25; 4.60	
Concentration 3 Nom; Meas (µg/L)	12.5; 18.3	
Concentration 4 Nom; Meas (µg/L)	24.9; 24.6	
Concentration 5 Nom; Meas (µg/L)	49.8; 44.0	
Concentration 6 Nom; Meas (µg/L)	99.5; 94.2	
Control	Negative: 0; 0	
	Solvent: 0; 0	
EC ₅₀ (95% CI) (μg/L)	28 (24.2-32.5)	Method: Weighted
		least squares
		nonlinear regression
NOEC	4.60	Method: ANOVA,
		Dunnett's test
		p:
		MSD:
% control at NOEC	98 %	5 d: 400,000 (tmt) /
		408,000 (mean
		controls) = 98

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires ASTM Type I water, and the medium is presumably appropriate for the test species because a specific culture media was used.

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2). Total: 100-2=98

<u>Acceptability:</u> Temperature variation (3), Random design (2), Minimum significant difference (1). Total: 100-6 = 94

Reliability score: mean(98,94)=96

Appendix A4 – Wildlife studies rated L

A. platyrhynchos. Emmerson 1978.

Documentation and acceptability rating for terrestrial laboratory/field data (adapted from ECOTOX 2006). Score is given if parameter is reported.

Parameter ¹	Score ²	Points
Exposure duration	20	20
Control type	7	7
Organism information (i.e., age, life stage)	8	8
Chemical grade or purity	5	0
Chemical analysis method	5	0
Exposure type (i.e., dermal, dietary, gavage)	10	10
Test location (i.e., laboratory, field, natural artificial)	5	5
Application frequency	5	5
Organism source	5	0
Organism number and/or sample number	5	5
Dose number Two concentrations plus control	5	0
Statistics		
Hypothesis tests		
Statistical significance	5	0
Significance level	5	0
Minimum significant difference	3	0
% of control at NOEC and/or LOEC Calculable at	3	3
highest concentration		
Point estimates (i.e., LC ₅₀ , EC ₅₀)	4	0
Total	100	63

^T Compiled from RIVM (2001), USEPA (1985; 2003b), ECOTOX (2006), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

² Weighting based acceptability criteria from various ASTM, OECD, APHA, and USEPA methods, ECOTOX (2006), and on data quality criteria in RIVM (2001), USEPA (1985; 2003b), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

A. platyrhynchos

Hudson, R.H., Tucker, R.K. and Haegele, M.A., 1984. Handbook of toxicity of pesticides to wildlife (No. 153). US Fish and Wildlife Service. USEPA MRID 160000.

Table X.x Documentation and acceptability rating for terrestrial laboratory/field data (adapted from ECOTOX 2006). Score is given if parameter is reported

parameter is reported.	g 2	
Parameter ¹	Score ²	Points
Exposure duration	20	20
Control type	7	7
Organism information (i.e., age, life stage)	8	8
Chemical grade or purity	5	5
Chemical analysis method	5	0
Exposure type (i.e., dermal, dietary, gavage)	10	10
Test location (i.e., laboratory, field, natural artificial)	5	5
Application frequency	5	5
Organism source	5	5
Organism number and/or sample number	5	0
Dose number	5	0
Statistics		
Hypothesis tests		
Statistical significance	5	0
Significance level	5	0
Minimum significant difference	3	0
% of control at NOEC and/or LOEC	3	0
Point estimates (i.e., LC ₅₀ , EC ₅₀)	4	0
Total	100	65

¹ Compiled from RIVM (2001), USEPA (1985; 2003b), ECOTOX (2006), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

² Weighting based acceptability criteria from various ASTM, OECD, APHA, and USEPA methods, ECOTOX (2006), and on data quality criteria in RIVM (2001), USEPA (1985; 2003b), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

Appendix A5 - Aqueous studies rated N

Asellus brevicaudus

Study: Sanders, HO. 1970. Toxicities of some herbicides to six species of freshwater crustaceans. *Journal of Water Pollution Control Federation*, 42, 1544-1550. EPA MRID 45088221.

RelevanceReliabilityScore: 70Score: 52.5Rating: LRating: N

EC₅₀ exceeds 2S so study automatically rates N and cannot be used in criteria calculation.

	Sanders 1970	A. brevicaudus
Parameter	Value	Comment
Test method cited	Previously described in peer review	Sanders HO and Cope OB. 1965. The relative toxicities of several pesticides to two spcies of Cladocerans. Transactions of the American Fisheries Society. 95, 165.
Phylum/subphylum	Arthropoda	
Class	Ispoda	
Order	Asellidae	
Family	Asellus	
Genus	brevicaudus	
Species	Arthropoda	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Not reported	
Source of organisms	Laboratory culture	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 96 h	
Effect 1	Mortality	
Control response 1	Not reported	
Temperature	21 ± 0.5 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	

	Sanders 1970	A. brevicaudus
Parameter	Value	Comment
Dilution water	Untreated well water	
pH	7.4	
Hardness	272 mg/L CaCO ₃	
Alkalinity	260 mg/L CaCO ₃	
Dissolved Oxygen	Not reported	Non-aerated water
Feeding	Not reported	
Purity of test substance	Technical	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	1.0 mL/L, methanol	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Concentrations, reps not	States that 4 or 5
	reported	concentrations and
		appropriate controls
		used
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	2000 (CI not reported)	Method: modified
	> 2S	Litchfield and
		Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Organism life stage/size (5), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8). Total: 100-32 =68

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Adequate organisms per rep (2), Feeding (3), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Number of concentrations (3), Random design (2), Adequate replication (2), Dilution factor (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-67 = 37

Reliability score: mean(68,37)=52.5

Bombina bombina

Study: Sayim F. (2010) Toxicity of trifluralin on the embryos and larvae of the red-bellied toad, *Bombina bombina. Turkish Journal of Zoology*, 34(4), pp.479-486.

LC/EC₅₀ exceeds 2S so study rates N and cannot be used for criteria derivation.

C. chroococcus sp.H4

Study: Aslim B, Ozturk S. (2009) Toxicity of herbicides to cyanobacterial isolates. *Journal of Environmental Biology*. 30(3), 381-384.

Test exposure concentrations all exceed 2S so study rates N and cannot be used for criteria derivation.

C. chroococcus

Study: Koksoy, H. and Aslim, B., 2013. Determination of Herbicide Resistance in Aquatic Cyanobacteria by Probit Analysis. Journal of Applied Biological Sciences, 7(2), pp.37-41.

 LC_{50} exceeds 2S so study rates N and cannot be used for criteria derivation.

Cypridopsis vidua

Study: Sanders, HO. 1970. Toxicities of some herbicides to six species of freshwater crustaceans. *Journal of Water Pollution Control Federation*, 42, 1544-1550. EPA MRID 45088221.

RelevanceReliabilityScore: 70Score: 52.5Rating: LRating: N

Relevance points taken off for: Endpoint (15), Controls (15). 100-30=70.

	Sanders 1970	C. vidua
Parameter	Value	Comment
Test method cited	Previously described in peer review	Sanders HO and Cope OB. 1965. The relative toxicities of several pesticides to two spcies of Cladocerans. Transactions of the American Fisheries Society. 95, 165.
Phylum/subphylum	Anthropoda	
Class	Ostracoda	
Order	Podocopida	
Family	Cyprididae	
Genus	Cypridopsis	
Species	Vidua	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Not reported	
Source of organisms	Laboratory culture	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 96 h	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	21 ± 0.5 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	

	Sanders 1970	C. vidua
Parameter	Value	Comment
Dilution water	Untreated well water	
pH	7.4	
Hardness	272 mg/L CaCO ₃	
Alkalinity	260 mg/L CaCO ₃	
Dissolved Oxygen	Not reported	Non-aerated water
Feeding	Not reported	
Purity of test substance	Technical	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	1.0 mL/L, methanol	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Concentrations, reps not	States that 4 or 5
	reported	concentrations and
		appropriate controls
		used
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	250 (CI not reported)	Method: modified
		Litchfield and
		Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Organism life stage/size (5), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8). Total: 100-32 =68

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Adequate organisms per rep (2), Feeding (3), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Number of concentrations (3), Random design (2), Adequate replication (2), Dilution factor (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-67 =37

Reliability score: mean(68,37)=52.5

Chlorella vulgaris

Study: Agirman N, Kendirlioglu G, Cetin AK. (2013) The effects of four pesticides on the growth of *Chlorella vulgais*. *Fresenius Environmental Bulletin*, 23(6), 1418-1422.

RelevanceReliabilityScore: 52.5Score: 54Rating: NRating: N

Relevance points taken off for: Standard method (15), Chemical purity (15), Toxicity value (15), Control response (7.5). 100-47.5 = 52.5

	Agirman et al. 2013	C. vulgaris
Parameter	Value	Comment
Test method cited	Not reported	
Division	Chlorophyta	
Class	Trebouxiophyceae	
Order	Chlorellales	
Family	Chlorellaceae	
Genus	Chlorella	
Species	vulgaris	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Algal cells	
Source of organisms	Isolated from plankton samples in a fishpond	
Have organisms been exposed to contaminants?	Not reported	
Animals acclimated and disease-free?	Not reported	
Animals randomized?	Not reported	Given organism size and presence in growth medium, it is assumed that aliquots are inherently randomly
Test vessels randomized?	Not reported	
Test duration	6 d	
Data for multiple times?	1, 2, 3, 4, 5, 6 d	
Effect 1	Cell count	
Control response 1	Not reported	
Temperature	23 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	16:8, 1:d/1000 lux	
Dilution water	Jaworski's growth medium	Made with distilled water

	Agirman et al. 2013	C. vulgaris
Parameter	Value	Comment
Feeding	Growth medium	
Purity of test substance	Not reported	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on nominal or measured concentrations?	Not reported	
Chemical method documented?	Not reported	
Concentration of carrier (if any) in test solutions	Not reported	
Concentration 1 Nom; Meas (µg/L)	1; not reported	3 reps, 10240 cells/mL
Concentration 2 Nom; Meas (µg/L)	3; not reported	3 reps, 10240 cells/mL
Concentration 3 Nom; Meas (µg/L)	7; not reported	3 reps, 10240 cells/mL
Concentration 4 Nom; Meas (µg/L)	10; not reported	3 reps, 10240 cells/mL
Concentration 5 Nom; Meas (µg/L)	15; not reported	3 reps, 10240 cells/mL
Concentration 6 Nom; Meas (µg/L)	20; not reported	3 reps, 10240 cells/mL
Control	0; 0	3 reps, 10240 cells/mL
EC ₅₀ (95% CI) (μg/L)	Not reported	Method: Not reported
NOEC	Not reported	Method: Not reported p: Not reported MSD: Not reported

Notes: Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires Type I water, and the medium is presumably appropriate for the test species because a specific culture media was used.

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Standard method (6), Chemical purity (5), Analytical method (4), Measured concentrations (3), Statistics method (5), Hypothesis tests (8), Point estimates (8). Total: 100-39 =61

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Chemical purity (10), Measured concentrations within 20% nominal (4), Carrier solvent (4), No prior contamination (4), Acclimation (1), Random design (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-53 =47

Reliability score: mean(47,61)=54

Daphnia magna

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

EC_{50} exceeds 2S so study rates N and cannot be used in criteria derivation.

	Johnson & Finley 1980	D. magna
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth phase	First instar	
Source of organisms	Federal or State hatchery Invertebrates collected from wild and cultured in laboratory	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	21 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized water	
рН	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	
Purity of test substance	95.9%	

	Johnson & Finley 1980	D. magna
Parameter	Value	Comment
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	560 (320-1000) > 2S	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation</u>: Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Daphnia magna

Study: Sanders, HO. 1970. Toxicities of some herbicides to six species of freshwater crustaceans. *Journal of Water Pollution Control Federation*, 42, 1544-1550. EPA MRID 45088221.

RelevanceReliabilityScore: 70Score: 52.5Rating: LRating: N

EC₅₀ exceeds 2S so study automatically rates N and cannot be used in criteria calculation.

	Sanders 1970	D. magna
Parameter	Value	Comment
Test method cited	Previously described in peer review	Sanders HO and Cope OB. 1965. The relative toxicities of several pesticides to two spcies of Cladocerans. Transactions of the American Fisheries Society. 95, 165.
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Not reported	
Source of organisms	Laboratory culture	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 96 h	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	21 ± 0.5 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	

	Sanders 1970	D. magna
Parameter	Value	Comment
Dilution water	Untreated well water	
pH	7.4	
Hardness	272 mg/L CaCO ₃	
Alkalinity	260 mg/L CaCO ₃	
Dissolved Oxygen	Not reported	Non-aerated water
Feeding	Not reported	
Purity of test substance	Technical	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	1.0 mL/L, methanol	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Concentrations, reps not	States that 4 or 5
	reported	concentrations and
		appropriate controls
		used
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	560 (CI not reported)	Method: modified
	> 2S	Litchfield and
		Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation</u>: Organism life stage/size (5), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8). Total: 100-32 =68

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Adequate organisms per rep (2), Feeding (3), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Number of concentrations (3), Random design (2), Adequate replication (2), Dilution factor (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-67 = 37

Reliability score: mean(68,37)=52.5

Daphnia pulex

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

EC_{50} exceeds 2S so study rates N and cannot be used in criteria derivation.

	Johnson & Finley 1980	D. pulex
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	pulex	
Family native to North America?	Yes	
Age/size at start of test/growth phase	First instar	
Source of organisms	Collected from wild and cultured in laboratory	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	15 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
рН	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	

	Johnson & Finley 1980	D. pulex
Parameter	Value	Comment
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	625 (446-876) > 2S	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Gambusia affinis

Study: Fabacher DL and Chambers H. (1974) Resistance to herbicides in insecticide-resistant mosquitofish, *Gambusia affinis*. *Environmental Letters* 7(1), 15-20.

 LC_{50} exceeds 2S so study rates N and cannot be used for criteria derivation.

Gammarus fasciatus

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

EC_{50} exceeds 2S so study rates N and cannot be used for criteria derivation.

	Johnson & Finley 1980	G. fasciatus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Anthropoda	
Class	Malacostraca	
Order	Amphipoda	
Family	Gammaridae	
Genus	Gammarus	
Species	fasciatus	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Mature	
Source of organisms	Invertebrates collected from wild and cultured in laboratory	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	21 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
pН	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	

	Johnson & Finley 1980	G. fasciatus
Parameter	Value	Comment
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	2200 (1400-3400) > 2S	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 = 55

Reliability score: mean(65,55)=60

Gammarus fasciatus

Study: Sanders, HO. 1970. Toxicities of some herbicides to six species of freshwater crustaceans. *Journal of Water Pollution Control Federation*, 42, 1544-1550. EPA MRID 45088221.

RelevanceReliabilityScore: 70Score: 52.5Rating: LRating: N

EC₅₀ exceeds 2S so study automatically rates N and cannot be used in criteria calculation.

	Sanders 1970	G. fasciatus
Parameter	Value	Comment
Test method cited	Previously described in peer review	Sanders HO and Cope OB. 1965. The relative toxicities of several pesticides to two spcies of Cladocerans. Transactions of the American Fisheries Society. 95, 165.
Phylum/subphylum	Anthropoda	
Class	Malacostraca	
Order	Amphipoda	
Family	Gammaridae	
Genus	Gammarus	
Species	fasciatus	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Not reported	
Source of organisms	Laboratory culture	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 96 h	
Effect 1	Mortality	
Control response 1	Not reported	
Temperature	21 ± 0.5 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	

	Sanders 1970	G. fasciatus
Parameter	Value	Comment
Dilution water	Untreated well water	
pH	7.4	
Hardness	272 mg/L CaCO ₃	
Alkalinity	260 mg/L CaCO ₃	
Dissolved Oxygen	Not reported	Non-aerated water
Feeding	Not reported	
Purity of test substance	Technical	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	1.0 mL/L, methanol	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Concentrations, reps not	States that 4 or 5
	reported	concentrations and
		appropriate controls
		used
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	1800 (CI not reported)	Method: modified
	> 2S	Litchfield and
		Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Organism life stage/size (5), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8). Total: 100-32 =68

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Adequate organisms per rep (2), Feeding (3), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Number of concentrations (3), Random design (2), Adequate replication (2), Dilution factor (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-67 = 37

Reliability score: mean(68,37)=52.5

Ictalurus punctatus

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

EC_{50} exceeds 2S so study rates N and cannot be used for criteria derivation.

	Johnson & Finley 1980	I. punctatus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Siluriformes	
Family	Ictaluridae	
Genus	Ictalurus	
Species	Punctatus	
Family native to North America?	Yes	
Age/size at start of test/growth	0.8	
phase		
Source of organisms	Hatchery	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	15 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
pН	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	

	Johnson & Finley 1980	I. punctatus
Parameter	Value	Comment
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	2200 (1420-3410)> 2S	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-45 = 55

Reliability score: mean(65,55)=60

Ictalurus punctatus

Study: McCorkle, F.M., Chambers, J.E. and Yarbrough, J.D., 1977. Acute toxicities of selected herbicides to fingerling channel catfish, *Ictalurus punctatus*. Bulletin of environmental contamination and toxicology, 18(3), pp.267-270.

Test exposure concentrations used all exceeded 2S so study rates N and cannot be used for criteria derivation.

Lepomis macrochirus

Study: No author, no date. Effect of trifluralin on bluegill sunfish fingerlings in aquaria tests. CA DPR 952909.

RelevanceReliabilityScore:Score:Rating:N

Relevance points taken off for: Standard (10), Chemical purity (15), Controls (15). 100-40=60.

	Geoffroy et al. 2003	L. macrochirus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Perciformes	
Family	Centrarchidae	
Genus	Lepomis	
Species	macrochirus	
Family native to North America?	Yes	
Age/size at start of test/growth	5 g	
phase		
Source of organisms	Not reported	
Have organisms been exposed to	Not reported	
contaminants?	-	
Animals acclimated and disease-	Not reported	
free?	-	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	No	
Effect 1	Mortality	
Control response 1	Not reported	
Temperature	24 °C	
Test type	Not reported	
Photoperiod/light intensity	Not reported	
Dilution water	Not reported	
рН	Not reported	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	
Feeding	Not reported	
Purity of test substance	Not reported	
Concentrations measured?	Not reported	

	Geoffroy et al. 2003	L. macrochirus
Parameter	Value	Comment
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	Acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	70; not reported	3 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	160; not reported	3 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	330; not reported	3 reps, 5/rep
Control	Not reported	
LC ₅₀ (95% CI) (μg/L)	89 (79.4-98.6)	Method: Probit

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Metacarcinus magister

Study: Caldwell RS. (1978) Biological effects of pesticides on the Dungeness crab. Environmental Research Laboratory, Office of Research and Development, US Environmental Protection Agency, Gulf Breeze, Florida.

RelevanceReliabilityScore: 67.5Score: 68.5Rating: NRating: L

Relevance points taken off for: Standard (10), Freshwater (15), Control response (7.5). 100-32.5=67.5

	Caldwell 1978	M. magister
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Antropoda/Crustacea	
Class	Malacostraca	
Order	Brachyura	
Family	Cancridae	
Genus	Metacarcinus	
Species	Magister	
Family native to North America?	Yes	
Age/size at start of test/growth	~2 y	
phase	80-100 mm	
Source of organisms	Yaquina Bay, Oregon	
Have organisms been exposed to	Not reported	
contaminants?		
Animals acclimated and disease-	Yes, 5 d	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	Acute: 96 h	
	Chronic, first: 85 d	
	Chronic, second: 90 d	
Data for multiple times?		
Effect 1	Mortality	
Control response 1		
Temperature	13 ± 1 °C	
Test type	Acute: Static renewal	
	Chronic: Flow-through	
Photoperiod/light intensity	Acute: 12:12, 1:d	
	Chronic, first: 11:13:, 1:d	
Dilution water	Natural seawater	Filtered, 25 ‰
		salinity
pН	7.5	

	Caldwell 1978	M. magister
Parameter	Value	Comment
Dissolved Oxygen	6.0 mg/L	Aerated
Feeding	Acute: not fed	
	Chronic, first: P. stellatus or	
	P. vetulus 3/w	
Purity of test substance	93 %	
Concentrations measured?	Yes	
Measured is what % of nominal?		
Toxicity values calculated based on		
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Acetone, 100 μL/L	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Acute: 30; not reported	0 reps, 10/rep
	Chronic: 1; 2.6	
Concentration 2 Nom; Meas (µg/L)	Acute: 93; not reported	
	Chronic: 10; 33	
Concentration 3 Nom; Meas (µg/L)	Acute: 300; not reported	
	Chronic: 100; 300	
Concentration 4 Nom; Meas (µg/L)	930; not reported >2S	
Concentration 5 Nom; Meas (µg/L)	3000; not reported $>2S$	
Concentration 6 Nom; Meas (µg/L)	9000; not reported > 2S	
Control	0;	
LC ₅₀ (95% CI) (μg/L)	>9300 (CI not reported) > 2S	Method: Straight
		line graphical
		interpolation
NOEC		Method:
		p:
		MSD:
LOEC		
MATC		
% control at NOEC		
24 1 1 2 5 5		
% control at LOEC		

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Standard method (6), Hardness (2), Alkalinity (2), Conductivity (2), Hypothesis tests (8), Point estimates (8). Total: 100- 28=72

<u>Acceptability:</u> Standard method (5), Control response (9), Measured concentrations within 20% nominal (4), No prior contamination (4), Organisms randomized (1), Hardness (2), Alkalinity (2), Conductivity (1), Random design (2), Adequate replication (2), Hypothesis tests (3). Total: 100-35 =65

Reliability score: mean(72,65)=68.5

Metacarcinus magister

Study: Caldwell RS. (1978) Biological effects of pesticides on the Dungeness crab. Environmental Research Laboratory, Office of Research and Development, US Environmental Protection Agency, Gulf Breeze, Florida.

RelevanceReliabilityScore: 67.5Score: 68.5Rating: NRating: L

Relevance points taken off for: Standard (10), Freshwater (15), Control response (7.5). 100-32.5=67.5

	Caldwell 1978	M. magister
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Antropoda/Crustacea	
Class	Malacostraca	
Order	Brachyura	
Family	Cancridae	
Genus	Metacarcinus	
Species	Magister	
Family native to North America?	Yes	
Age/size at start of test/growth	Eggs	
phase		
Source of organisms	Collected from single	
	wildcaught female	
Have organisms been exposed to	Not reported	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	24 h	
Data for multiple times?	No	
Effect 1	Egg hatching	
Control response 1	Not reported	
Temperature	13 ± 1 °C	
Test type	Static renewal	
Photoperiod/light intensity	Not reported	
Dilution water	Natural seawater	25 % salinity
рН	Not reported	
Hardness	mg/L CaCO ₃	
Alkalinity	mg/L CaCO ₃	
Conductivity	μS/cm	
Dissolved Oxygen	Air saturation	

	Caldwell 1978	M. magister
Parameter	Value	Comment
Feeding	Not reported	
Purity of test substance	93 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations? Chemical method documented?	GC	
Concentration of carrier (if any) in test solutions	Acetone, 100 μL/L	
Concentration 1 Nom; Meas (µg/L)	0.0033; Not reported	2 reps, 30/rep
Concentration 2 Nom; Meas (µg/L)	0.010; Not reported	
Concentration 3 Nom; Meas (µg/L)	0.033; Not reported	
Concentration 4 Nom; Meas (µg/L)	0.10; Not reported	
Concentration 5 Nom; Meas (µg/L)	0.33; Not reported	
Control	0; 0	
LC ₅₀ (95% CI) (μg/L)	Not reported	Method: Straight line graphical interpolation
NOEC	>330	Method: p: MSD:

Trifluralin solubility (S) = $238.08 \mu g/L$, $2S = 476.16 \mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Standard method (6), Hardness (2), Alkalinity (2), Conductivity (2), Hypothesis tests (8), Point estimates (8). Total: 100-28=72

<u>Acceptability:</u> Standard method (5), Control response (9), Measured concentrations within 20% nominal (4), No prior contamination (4), Organisms randomized (1), Hardness (2), Alkalinity (2), Conductivity (1), Random design (2), Adequate replication (2), Hypothesis tests (3). Total: 100-35 =65

Reliability score: mean(72,65)=68.5

Metacarcinus magister

Study: Caldwell RS. (1978) Biological effects of pesticides on the Dungeness crab. Environmental Research Laboratory, Office of Research and Development, US Environmental Protection Agency, Gulf Breeze, Florida.

RelevanceReliabilityScore: 67.5Score: 68.5Rating: NRating: L

Relevance points taken off for: Standard (10), Freshwater (15), Control response (7.5). 100-32.5=67.5

	Caldwell 1978	M. magister
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Antropoda/Crustacea	
Class	Malacostraca	
Order	Brachyura	
Family	Cancridae	
Genus	Metacarcinus	
Species	Magister	
Family native to North America?	Yes	
Age/size at start of test/growth	96 h bioassay: First instar	
phase	Chronic bioassay: 2	
	months/3 rd instar	
Source of organisms	Yaquina Bay, Oregon	
Have organisms been exposed to	Not reported	
contaminants?		
Animals acclimated and disease-	Chronic, second: 2 months	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	Acute: 96 h	
	Chronic, first: 36 d	
	Chronic: second: 80 d	
Data for multiple times?	Not reported	
Effect 1	Mortality	
Control response 1	Not reported	
Effect 2	Inability to right from	
	overturned position	
Control response 2	Not reported	
Temperature	13 ± 1 °C	
Test type	Acute: Static renewal	
	Chronic: Flow-through	
Photoperiod/light intensity	15:9, 1:d	

	Caldwell 1978	M. magister
Parameter	Value	Comment
Dilution water	Natural seawater	Filtered, UV sterilized Acute: 25 % salinity Chronic: 31-34 % salinity
рН	7.3-8.1	-
Dissolved Oxygen	>5.5 mg/L	
Feeding	Acute: not fed Chronic: Cockle clams, rockfish weekly	
Purity of test substance	93 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	126-173 %	
Toxicity values calculated based on nominal or measured concentrations?	Not reported	
Chemical method documented?	GC	
Concentration of carrier (if any) in test solutions	Acetone, 100 μL/L	
Concentration 1 Nom; Meas (µg/L)	Acute: 3.3; not reported Chronic, first: 0.15; not reported	0 reps, 10/rep
Concentration 2 Nom; Meas (µg/L)	Acute: 10; not reported Chronic, first: 1.5; 2.6	
Concentration 3 Nom; Meas (µg/L)	Acute: 33; not reported Chronic, first: 15; 19	
Concentration 4 Nom; Meas (µg/L)	Acute: 100; not reported Chronic: 150; 190	
Concentration 5 Nom; Meas (µg/L)	Acute: 330; not reported	
Concentration 6 Nom; Meas (µg/L)	Acute: 1000; not reported >2S	
Control	0; 0	
LC ₅₀ (95% CI) (μg/L)	>1000 (CI not reported) >2S	Method: Straight line graphical interpolation
EC ₅₀ (95% CI) (μg/L)	>1000 (CI not reported) >2S	Method: Straight line graphical interpolation

Notes: Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Standard method (6), Hardness (2), Alkalinity (2), Conductivity (2), Hypothesis tests (8), Point estimates (8). Total: 100- 28=72

<u>Acceptability:</u> Standard method (5), Control response (9), Measured concentrations within 20% nominal (4), No prior contamination (4), Organisms randomized (1), Hardness (2), Alkalinity

(2), Conductivity (1), Random design (2), Adequate replication (2), Hypothesis tests (3). Total: 100-35=65

Reliability score: mean(72,65)=68.5

Metacarcinus magister

Study: Caldwell RS. (1978) Biological effects of pesticides on the Dungeness crab. Environmental Research Laboratory, Office of Research and Development, US Environmental Protection Agency, Gulf Breeze, Florida.

RelevanceReliabilityScore: 67.5Score: 68.5Rating: NRating: L

Relevance points taken off for: Standard (10), Freshwater (15), Control response (7.5). 100-32.5=67.5

	Caldwell 1978	M. magister
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Antropoda/Crustacea	
Class	Malacostraca	
Order	Brachyura	
Family	Cancridae	
Genus	Metacarcinus	
Species	Magister	
Family native to North America?	Yes	
Age/size at start of test/growth	Eggs	
phase		
Source of organisms	Wildcaught of Newport	
Have organisms been exposed to	Not reported	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	Chronic: 69 d	
Data for multiple times?	Not reported	
Effect 1	Mortality	
Control response 1	~ <15 %	
Effect 2	Motility	
Control response 2	100%	
Temperature	13 ± 1 °C	
Test type	Acute: Static renewal	
	Chronic: Flow-through	
Photoperiod/light intensity	Acute: 9:15, 1:d	
	Chronic: 12.5:10.5, 1:d	
Dilution water	Natural seawater	Filtered, UV
		sterilized, 25 ‰
		salinity

	Caldwell 1978	M. magister
Parameter	Value	Comment
pH	7.8	
Hardness	mg/L CaCO ₃	
Alkalinity	mg/L CaCO ₃	
Conductivity	μS/cm	
Dissolved Oxygen	Acute: >7.0 mg/L	
	Chronic: >8 mg/L	
Feeding	Acute: Not reported	
	Chronic: live Artemia salina	
	2-3/w	
Purity of test substance	93 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	273-353 %	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?	CC	
Chemical method documented?	GC	
Concentration of carrier (if any) in test solutions	Acetone, 100 μL/L	
	Acute: 0.35; not reported	0 reps, 20/rep
Concentration 1 Nom; Meas (µg/L)	Chronic, first test: 0.15; not	0 1eps, 20/1ep
	reported	
	Chronic, second test: 1.5;	
	3.1	
Concentration 2 Nom; Meas (µg/L)	Acute: 1.1; not reported	
Concentration 2 1 (om, 172as (µg/2)	Chronic, first test: 1.5; 4.1	
	Chronic, second test: 15; 26	
Concentration 3 Nom; Meas (µg/L)	Acute: 3.5; not reported	
	Chronic, first test: 15; 53	
	Chronic, second test:150;	
	220	
Concentration 4 Nom; Meas (µg/L)	Acute: 11; not reported	
	Chronic, first test: 150; 480	
Concentration 5 Nom; Meas (µg/L)	Acute: 35; not reported	
Concentration 6 Nom; Meas (µg/L)	Acute: 110; not reported	
Control	0; 0	8 reps, 20/rep
LC ₅₀ (95% CI) (μg/L)	Acute: >110 (CI not	Method: Straight
	reported)	line graphical
	(O) (GI	interpolation
EC ₅₀ (95% CI) (μg/L)	Acute: 60 (CI not reported)	Method:
NOEC	Not reported	Method: not
		reported
		p: not reported
Notes:		MSD: not reported

Trifluralin solubility (S) = 238.08 $\mu g/L$, 2S = 476.16 $\mu g/L$.

Reliability points taken off for:

<u>Documentation:</u> Standard method (6), Hardness (2), Alkalinity (2), Conductivity (2), Hypothesis tests (8), Point estimates (8). Total: 100- 28=72

<u>Acceptability:</u> Standard method (5), Control response (9), Measured concentrations within 20% nominal (4), No prior contamination (4), Organisms randomized (1), Hardness (2), Alkalinity (2), Conductivity (1), Random design (2), Adequate replication (2), Hypothesis tests (3). Total: 100-35 =65

Reliability score: mean(72,65)=68.5

Orconectes nais

Study: Sanders, HO. 1970. Toxicities of some herbicides to six species of freshwater crustaceans. *Journal of Water Pollution Control Federation*, 42, 1544-1550. EPA MRID 45088221.

RelevanceReliabilityScore: 70Score: 52.5Rating: LRating: N

EC₅₀ exceeds 2S so study automatically rates N and cannot be used in criteria calculation.

	Sanders 1970	O. nais
Parameter	Value	Comment
Test method cited	Previously described in peer review	Sanders HO and Cope OB. 1965. The relative toxicities of several pesticides to two spcies of Cladocerans. Transactions of the American Fisheries Society. 95, 165.
Phylum/subphylum	Arthropoda	
Class	Malacostraca	
Order	Decapoda	
Family	Cambaridae	
Genus	Orconectes	
Species	Nais	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Not reported	
Source of organisms	Laboratory culture	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 96 h	
Effect 1	Mortality	
Control response 1	Not reported	
Temperature	21 ± 0.5 °C	
Test type	Static	

	Sanders 1970	O. nais
Parameter	Value	Comment
Photoperiod/light intensity	Not reported	
Dilution water	Untreated well water	
pH	7.4	
Hardness	272 mg/L CaCO ₃	
Alkalinity	260 mg/L CaCO ₃	
Dissolved Oxygen	Not reported	Non-aerated water
Feeding	Not reported	
Purity of test substance	Technical	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	1.0 mL/L, methanol	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Concentrations, reps not	States that 4 or 5
	reported	concentrations and
		appropriate controls
		used
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	50000 (CI not reported)	Method: modified
	> 2S	Litchfield and
		Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Organism life stage/size (5), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8). Total: 100-32 =68

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Adequate organisms per rep (2), Feeding (3), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Number of concentrations (3), Random design (2), Adequate replication (2), Dilution factor (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-67 =37

Reliability score: mean(68,37)=52.5

Pteronarcys californica

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

EC_{50} exceeds 2S so study rates N and cannot be used for criteria derivation.

	Johnson & Finley 1980	P. californica
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Arthropoda	
Class	Insecta	
Order	Plecoptera	
Family	Pteronarcyidae	
Genus	Pteronarcys	
Species	Californica	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Second year class	
Source of organisms	Invertebrates collected from wild and cultured in laboratory	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	15 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized	
	water	
pН	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	
Purity of test substance	95.9 %	
Concentrations measured?	Not reported	

	Johnson & Finley 1980	P. californica
Parameter	Value	Comment
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	2800 (2100-3700) > 2S	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation</u>: Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60

Procambarus clarkii

Study: Naqvi, S.M. and Leung, T.S., 1983. Trifluralin and oryzalin herbicides toxicities to juvenile crawfish (*Procambarus clarkii*) and mosquitofish (*Gambusia affinis*). Bulletin of environmental contamination and toxicology, 31(3), pp.304-308.

Test exposure concentrations all exceed 2S so study rates N and cannot be used for criteria derivation.

Palaemonetes kadiakensis

Study: Sanders, HO. 1970. Toxicities of some herbicides to six species of freshwater crustaceans. *Journal of Water Pollution Control Federation*, 42, 1544-1550. EPA MRIDs 45088221 and 5001497.

RelevanceReliabilityScore: 70Score: 52.5Rating: LRating: N

EC₅₀ exceeds 2S so study automatically rates N and cannot be used in criteria calculation.

	Sanders 1970	P. kadiakensis
Parameter	Value	Comment
Test method cited	Previously described in peer review	Sanders HO and Cope OB. 1965. The relative toxicities of several pesticides to two spcies of Cladocerans. Transactions of the American Fisheries Society. 95, 165.
Phylum/subphylum	Arthropoda	
Class	Malacostraca	
Order	Decapoda	
Family	Palaemonidae	
Genus	Palaemonetes	
Species	Kadiakensis	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Not reported	
Source of organisms	Laboratory culture	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 96 h	
Effect 1	Mortality	
Control response 1	Not reported	
Temperature	21 ± 0.5 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	

	Sanders 1970	P. kadiakensis
Parameter	Value	Comment
Dilution water	Untreated well water	
pH	7.4	
Hardness	272 mg/L CaCO ₃	
Alkalinity	260 mg/L CaCO ₃	
Dissolved Oxygen	Not reported	Non-aerated water
Feeding	Not reported	
Purity of test substance	Technical	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	1.0 mL/L, methanol	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Concentrations, reps not	States that 4 or 5
	reported	concentrations and
		appropriate controls
		used
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	1200 (CI not reported)	Method: modified
	> 2S	Litchfield and
		Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation:</u> Organism life stage/size (5), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8). Total: 100-32 =68

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Adequate organisms per rep (2), Feeding (3), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Number of concentrations (3), Random design (2), Adequate replication (2), Dilution factor (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-67 = 37

Reliability score: mean(68,37)=52.5

Proisotoma minuta

Study: Park, E.K. and Lees, E.M., 2005. Application of an artificial sea salt solution to determine acute toxicity of herbicides to Proisotoma minuta (Collembola). Journal of Environmental Science and Health Part B, 40(4), pp.595-604.

Test exposure concentrations all exceed 2S so study rates N and cannot be used for criteria derivation.

Palaemonetes pugio

Study: Adams ER and Grothe DW. (1988) Acute toxicity of trifluralin to the grass shrimp (*Palaemonetes pugio*) in a flow-through test system. Lilly Research Laboratories, Greenfield, Indiana. Laboratory project identification C01687. USEPA MRID 40674801.

 LC_{50} of 638 $\mu g/L$ exceeds 2S so study rates N and cannot be used for criteria derivation.

R. subcapitata

Study: Ordog V and Kuivasniemi K. (1989) Studies on the Effect of Cell Division-inhibiting Herbicides on Unialgal and Mixed Algal Cultures. *Internationale Revue der gesamten Hydrobiologie und Hydrographie*, 74(2), 221-226.

EC₅₀ exceeds 2S so study rates N and cannot be used for criteria derivation.

Simocephalus serrulatus

Study: Johnson WW and Finley MT. (1980) U.S. Department of Interior, Fish and Wildlife Service. *Handbook of Acute Toxicity of Chemicals to Fish and Aquatic Invertebrates*. Resource Publication No. 137. Washington, DC: U.S. Government Printing Office.

EC_{50} exceeds 2S so study rates N and cannot be used in criteria derivation.

	Johnson & Finley 1980	S. serrulatus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Phyllopoda	
Family	Diplostraca	
Genus	Simocephalus	
Species	Serrulatus	
Family native to North America?	Yes	
Age/size at start of test/growth phase	First instar	
Source of organisms	Federal or State hatchery Invertebrates collected from wild and cultured in laboratory	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	Not reported	
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	15 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Reconstituted deionized water	
рН	7.2-7.5	
Hardness	40-50 mg/L CaCO ₃	
Alkalinity	30-35 mg/L CaCO ₃	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Aerated beforehand
Feeding	Not fed	
Purity of test substance	95.9%	

	Johnson & Finley 1980	S. serrulatus
Parameter	Value	Comment
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	≤0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	≥6 concentrations tested but	2 reps, 10/rep
	not reported	
Control	Not reported	
EC ₅₀ (95% CI) (μg/L)	900 (651-1245) > 2S	Method: Litchfield
		and Wilcoxon

Trifluralin solubility (S) = 238.08 μ g/L, 2S = 476.16 μ g/L.

Reliability points taken off for:

<u>Documentation</u>: Control type (8), Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100-35 =65

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Dissolved oxygen (6), Conductivity (1), Photoperiod (2), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100-45 =55

Reliability score: mean(65,55)=60